Synthesis of Cyclohexapeptides containing Pro and Aib residues

Jeremic, Tatjana ; Linden, Anthony ; Heimgartner, Heinz

Abstract: Cyclization reactions on hexapeptides containing several alpha-aminoisobutyric acid (= 2-amino-2-methylpropanoic acid; Aib) residues and the turn-promoting glycine (Gly) and proline (Pro) residues were investigated. Eight linear hexapeptides were synthesized, and their cyclization was attempted with various coupling reagents. The macrolactamization step proved to be difficult since only three hexapeptides could be cyclized. Two of these latter peptides were the linear precursors of the same cyclic hexapeptide, cyclo(Aib-Aib-Phe-Pro-Aib-Gly) (1). Surprisingly, they gave 1 in almost the same yield. Thus, 1 was obtained in 35% yield upon ring closure at the Phe/Pro site by using DEPBT as the coupling reagent, whereas the cyclization at the Aib/Phe site led to 1 in 28 and 34% yield by using PyAOP and DEPC, respectively (DEPBT = 3-[(diethoxyphosphoryl)oxy]-1,2,3-benzotriazin-4(3H)-one, PyAOP = (1H-7-azabenzotriazol-1-yloxy)tripyrroli- din-1-ylphosphonium hexafluorophosphate, DEPC = diethyl phosphorocyanidate). Another cyclic hexapeptide, cyclo(Aib-Aib-Gly-Aib-Pro-Gly) (2) was prepared in 34% yield when DEPC was used in the cyclization step. The solid-state conformation of 1 was established by X-ray crystallography.

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Prof. Dr. H. Heimgartner

Tel.: 01 635 42 82

Fax: 01 635 68 12

e-mail: heimgart@oci.unizh.ch

Synthesis of Cyclohexapeptides Containing Pro- and Aib-Residues

by Tatjana Jeremic[1]), Anthony Linden, and Heinz Heimgartner*

Organisch-chemisches Institut der Universität Zürich, Winterthurerstrasse 190,

CH-8057 Zürich

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Cyclization reactions on hexapeptides containing several Aib residues and the turn promoting Gly and Pro residues were investigated. Eight linear hexapeptides were synthesized and their cyclization was attempted using various coupling reagents. The macrolactamization step proved to be difficult since only three of these hexapeptides could be cyclized. Two of these peptides were the linear precursors of the same cyclic hexapeptide, *cyclo*(Pro-Aib-Gly-Aib-Aib-Phe) (**1**). Surprisingly, they gave **1** in almost the same yield. Thus, **1** was obtained in 35% yield upon ring closure at the Phe/Pro site using DEPBT as the coupling reagent, whereas the cyclization at the Aib/Phe site led to **1** in 28 and 34% yield by using PyAOP and DEPC, respectively. Another cyclic hexapeptide, *cyclo*(Gly-Aib-Pro-Gly-Aib-Aib) (**2**) was prepared in 34% yield when DEPC was used in the cyclization step. The solid-state conformation of **1** was established by X-ray crystallography.

1. Introduction. – Cyclic peptides usually exist in more clearly defined conformations than their linear counterparts, and often have increased receptor affinity and metabolic stability. Out of this arises their potential to serve as lead structures in drug design [1]. Cyclization can also constrain a short amino acid sequence to a turn conformation [2]. In particular, cyclic penta- and hexapeptides have frequently been used as models for reverse turns, although the cyclization of these small peptides is often troublesome [3]. Since peptide bonds possess pronounced π character and preferentially adopt a *trans* conformation, the linear precursor has the terminal acid and amine functions in remote positions, which is unfavorable for cyclization. Incorporation of residues such as Gly, Pro or D-amino acids are known to enhance cyclization yields [4].

Recently, we have focused our interest on the cyclization of model hexapeptides containing Aib (α-aminoisobutyric acid) residues, and we were also interested in the propensity of these cyclic peptides to form certain types of β-turns. Since we have successfully cyclized hexapeptides containing several Aib residues and one or two Gly residues [5], the aim of this work was to investigate the cyclization tendencies of hexapeptides containing Aib and Pro residues. Pro is believed to prefer turn-forming *cis*-peptide bonds, which should facilitate the ring closure. In this paper we report the synthesis of *cyclo*(Pro-Aib-Gly-Aib-Aib-Phe) (**1**) and *cyclo*(Gly-Aib-Pro-Gly-Aib-Aib) (**2**) and the attempted cyclizations of four additional hexapeptides containing Aib and Pro residues. Furthermore, three different linear precursors of the cyclic peptide **1** were synthesized in order to study the influence of the amino acid sequence on the cyclization yield.

Formulae

2. Results and Discussion. – 2.1. *Preparation of Linear Peptides and their Cyclization*. The amino acid sequence of the linear precursor and the choice of the coupling reagent are two of the most important factors that affect the result of the cyclization reaction [6] [7]. The cyclic hexapeptide *cyclo*(Pro-Aib-Gly-Aib-Aib-Phe) (**1**) was chosen as a model for the investigation of the significance of the linear amino acid sequence on the success of the macrolactamization. In the retrosynthetic analysis, three amide bonds were selected for the final ring closure step, *i.e.* between Phe/Pro, Aib/Phe and Aib/Gly, respectively. These three disconnections were likely to give different yields for the cyclization, but it was impossible to predict with appropriate certainty which one would be optimal.

Scheme 1

The synthesis of the linear precursor for the Phe/Pro cyclization site was carried out by stepwise elongation from the *N*-terminal Pro derivative (*Scheme 1*). The synthesis started by coupling Fmoc-Pro-OH with the 2*H*-azirin-3-amine **3**, our synthon for Aib, to give the dipeptide amide **4**, which was then hydrolyzed with 3N HCl (MeCN/H_2O 1:1) to give the dipeptide acid Fmoc-Pro-Aib-OH (**5**). The *N*-terminal tripeptide Z-Gly-Aib-Aib-N(Me)Ph (**6**) was prepared analogously from Z-Gly-OH by use of the 'azirine/oxazolon method' [8]. After removal of the Z group from **6**, PyBOP-mediated[2]) coupling with **5** gave pentapeptide amide **7** in

[2]) For abbreviations see *Exper. Part., Section 1.*

69% yield. The latter was hydrolyzed to yield the peptide acid **8**, which was then coupled with H-Phe-O*t*Bu to give the linear precursor Fmoc-Pro-Aib-Gly-Aib-Aib-Phe-O*t*Bu (**9**) in high yield. The coupling reaction was again accomplished by use of PyBOP. Finally, after removal of the Fmoc group at the *N*-terminus of **9** by treatment with Et_2NH and then deprotection of the *t*-butyl ester group at the *C*-terminus with TFA, the free linear precursor as its TFA salt was subjected to macrolactamization.

The cyclic hexapeptide **1** was initially obtained in low yield (20%) by ring closure with TBTU/HOBt/DIEA[2]), however, due to the long reaction time of 6 d, significant epimerization of the *C*-terminal Phe residue occurred, and afforded **1** as an inseparable mixture of two diastereoisomers. For this reason, the DEPBT[2]) coupling reagent was chosen to be used in the cyclization reaction. This reagent has proven to be very efficient in the synthesis of cyclic peptides [9] [10] with a remarkable resistance to racemization [11]. The cyclization was carried out with three equivalents of DEPBT in THF and high dilution (1.5 mM) and in the presence of DIEA during 3 d. This procedure afforded *cyclo*(Pro-Aib-Gly-Aib-Aib-Phe) (**1**) in 35% yield. No side-reactions, such as dimerization of the linear precursor or epimerization of the *C*-terminal Phe residue were observed, which allowed the desired cyclic peptide **1** to be purified easily.

The cyclization at the Aib/Phe site should be advantageous, because epimerization is not possible when Aib is the *C*-terminal residue, and the steric hindrance should be reduced compared with that of the Phe/Pro cyclization site. Therefore, the linear precursor for this cyclization was prepared as shown in *Scheme 2*. The dipeptide amide Fmoc-Pro-Aib-N(Me)Ph (**4**) was first *N*-

deprotected (Et_2NH in CH_2Cl_2) and then coupled with Z-Phe-OH using PyAOP[2]), which lead to the tripeptide Z-Phe-Pro-Aib-N(Me)Ph (**10**) in high yield. The hydrolysis of **10** with 3N HCl (MeCN/H_2O 1:1) provided tripeptide acid **11**. After deprotection of the N-terminus of Z-Gly-Aib-Aib-O*t*Bu (**12**), the reaction with **11** using HATU/HOAt as activating agents afforded hexapeptide Z-Phe-Pro-Aib-Gly-Aib-Aib-O*t*Bu (**13**) in good yield. After the removal of the Z group of **13** and subsequent cleavage of the *tert*-butyl group by TFA, the linear precursor was cyclized using different coupling reagents. The best result among the reagents explored was achieved when DEPC ($(EtO)_2P(O)CN$) [12] was used. The cyclic hexapeptide **1** was obtained in 34% yield when a diluted solution (1.0 mM) of the deprotected hexapeptide in DMF was exposed to DEPC and DIEA for 6 d at room temperature. It should be noted that the yield of the crude cyclic product was higher when PyAOP/HOAt was used as the cyclization reagent, and the reaction was faster (24 h), but the purification was difficult, which resulted in a slightly lower yield (28%) of pure **1**. An additional fraction of **1**, contaminated with some $Et(i\text{-}Pr)_2NH^+PF_6^-$, was obtained but could not be purified. We were unable to completely remove this byproduct by washing with aqueous citric acid and $NaHCO_3$ nor after two successive chromatographic purifications (AcOEt/MeOH 10:1, followed by CH_2Cl_2/MeOH 15:1) or crystallization. The only singlecrystals, suitable for X-ray crystallographic analysis, that could be grown from this mixture were co-crystals of $Et(i\text{-}Pr)_2NH^+PF_6^-$ and **1**. The formation of **1** was also attempted using the DEPBT coupling reagent, but surprisingly the linear precursor remained unaltered after a period of 3 d.

Scheme 2

The linear precursor for the cyclization at the Aib/Gly site was prepared as outlined in *Scheme 4*. After removal of the Z group in Z-Phe-Pro-Aib-N(Me)Ph (**10**), condensation with Z-Gly-Aib-Aib-OH using PyAOP/HOAt provided the linear hexapeptide **14** in 90% yield. The deprotection of **14** afforded the free linear precursor, which was subjected to macrolactamization with four different coupling reagents, *i.e.*, DEPC, DEPBT, PyAOP/HOAt, and HATU/HOAt. Unfortunately, all four attempts failed and only decomposition of the coupling reagents was observed. This result was very surprising since we have previously successfully cyclized several Aib-containing hexapeptides at an Aib/Gly site in good yields (45-57%) [5] [13]. We assume that the Pro residue at position 5 of the sequence makes the peptide more rigid, thereby completely preventing cyclization.

Scheme 3

The incorporation of Gly or Pro residues as turn-inducing elements is generally considered as a prerequisite for the ring closure of short peptides [14]. Since the cyclic peptide **1** contains both of these residues, the cyclization was expected to proceed in high yield. The observed moderate yields (28-35%) were presumably caused by the presence of three conformationally constrained Aib units in the sequence, in addition to one conformationally restricted Pro residue, thus evoking highly constrained linear precursors. Based on these precedents, we

decided to prepare a similar cyclohexapeptide, *i.e. cyclo*(Gly-Aib-Pro-Gly-Aib-Aib) (**2**), which contains an additional Gly instead of Phe. Furthermore, one Gly residue is placed adjacent to the Pro unit, which should hopefully decrease the conformational constraint and enhance the cyclization yield. The synthesis of the linear precursor of **2** and its cyclization are shown in *Scheme 4*. After removal of the Z group of tripeptide **6**, condensation with Fmoc-Pro-OH using PyAOP yielded the linear tetrapeptide Fmoc-Pro-Gly-Aib-Aib-N(Me)Ph (**15**). Treatment of **15** with Et_2NH gave the *N*-deprotected tetrapeptide, which on subsequent coupling with Z-Gly-Aib-OH using PyAOP led to the linear hexapeptide Z-Gly-Aib-Pro-Gly-Aib-Aib-N(Me)Ph (**16**). The latter was hydrolyzed to give the peptide acid **17**, the Z group was removed hydrogenolytically, and the free linear precursor subjected to cyclization in DMF (1.5 mM) by using DEPC (4 equiv.) as the coupling reagent in the presence of DIEA. The cyclopeptide **2** was obtained as a white precipitate, which was almost insoluble in organic solvents and H_2O. Since chromatographic purification was not possible, **2** was collected by filtration, washed with H_2O, MeOH and CH_2Cl_2 to remove excess coupling reagent and some byproducts and was recrystallized from hot H_2O/MeOH/EtOH solution. In this way, pure cyclohexapeptide **2** was obtained in 34% yield[3]).

[3]) The cyclization was not repeated using other coupling reagents such as PyAOP, DEPBT or HATU because, in contrast to DEPC, they are solid materials and it would not be possible to remove them from the cyclic product merely by filtration and washing. In addition, they usually generate much more of the byproducts during cyclization or work up, with some byproducts arising from the decomposition of excess coupling reagent.

Due to the low solubility of **2**, the NMR spectra had to be performed in DMSO containing small amounts of TFA. Surprisingly, although the linear precursor of **2** possesses the turn-inducing Pro-Gly segment in the middle of the sequence and two residues (Aib and Gly) at the C- and N-terminus, which can both be considered as pseudo-D- and L-amino acids, the cyclization was no more effective than in the case of **1**.

Scheme 4

Recently, we have synthesized a cyclic hexapeptide in which Aib residues alternate with proteinogenic amino acids, *i.e.*, *cyclo*(Gly-Aib-Leu-Aib-Phe-Aib) [15]. The conformation of this cyclic peptide in solution was well defined and it was shown that it is very similar to the conformation adopted in the solid state. Furthermore, the cyclization yield was surprisingly high (53%), although the Aib unit situated between the two large Leu and Phe residues was forced to assume a fully extended conformation, as shown by X-ray crystallography and MD simulations. It is unfavorable and highly uncommon for an Aib residue to adopt extended conformations. Thus, we decided to replace Leu by Pro in order to see how this modification would affect the cyclization yield and the conformational preference of the Aib residues. The synthesis of the linear precursor is outlined in *Scheme 5*. After Fmoc deprotection of dipeptide amide **4**, the coupling with Z-Gly-Aib-OH using the PyAOP reagent gave tetrapeptide Z-Gly-Aib-Pro-Aib-N(Me)Ph (**18**) in high yield. The latter was hydrolyzed (3N HCl (MeCN/H_2O 1:1)) to give peptide acid **19**. The Z group of Z-Phe-Aib-O*t*Bu (**20**) was removed,

and the subsequent coupling with **19** by using of PyAOP provided hexapeptide Z-Gly-Aib-Pro-Aib-Phe-Aib-O*t*Bu (**21**) in moderate yield. After the removal of the protecting groups, the linear precursor was subjected to cyclization, but both DEPBT and PyBOP, failed to give *cyclo*(Gly-Aib-Pro-Aib-Phe-Aib) (**22**). Unfortunately, we were unable to grow crystals of the linear hexapeptide suitable for an X-ray crystal structure analysis in order to evaluate the conformational constraints of the linear precursor.

Scheme 5

Scheme 6

Next, we prepared a similar hexapeptide, with Pro at the *N*-terminus and the Gly residue in the middle of the sequence, with the aim of facilitating the cyclization (*Scheme 6*). Starting with the dipeptide amide **23** and Fmoc-Pro-Aib-OH (**5**), and using HATU/HOAt as the coupling reagent, the tetrapeptide amide Fmoc-Pro-Aib-Gly-Aib-N(Me)Ph (**24**) was obtained and hydrolyzed to give **25**. Coupling of H-Phe-Aib-N(Me)Ph, generated by hydrogenolysis from Z-Phe-Aib-N(Me)Ph (**26**) [16] with **25** by using TBTU/HOBt afforded Fmoc-Pro-Aib-Gly-Aib-Phe-Aib-N(Me)Ph (**27**). After deprotection, the linear precursor was subjected to cyclization. Unfortunately, both PyAOP and DEPBT did not lead to *cyclo*(Pro-Aib-Gly-Aib-Phe-Aib) (**28**). Since the linear precursor of **28**, as in the case of **22**, remained unaltered upon exposure to the coupling reagents and the

base over a period of several days, we suppose that these hexapeptide sequences form rigid extended conformations which prevent cyclization.

Scheme 7

The cyclic Pro- and Aib-containing pentapeptide *cyclo*(Gly-Aib-Pro-Aib-Aib) has previously been synthesized in excellent yield (89%) using DEPC/DIEA in DMF in an overnight reaction [16]. The linear precursor of this cyclic pentapeptide apparently possess a cyclization-prone conformation in DMF solution. Although it is known that minor changes in peptide sequence can significantly affect cyclization, we hoped that by incorporation of an additional Aib residue into the above sequence, the conformational preference of the linear precursor would not be altered too much and would, therefore, lead to the cyclic hexapeptide in good yield. The synthesis of the linear precursor containing Gly, Pro and four Aib residues is outlined in *Scheme 7*. PyBOP-mediated condensation of Fmoc-Pro-Aib-OH (**5**) with H-Aib-O*t*Bu gave tripeptide Fmoc-Pro-Aib-Aib-O*t*Bu (**29**) in good yield. Removal of the Fmoc group of **29** followed by a second PyBOP-mediated coupling with Z-Gly-Aib-Aib-OH provided Z-Gly-Aib-Aib-Pro-Aib-Aib-O*t*Bu (**30**) in moderate yield. Finally, the protecting groups were removed and the linear precursor was subjected to cyclization. However, all attempts using either DEPBT/DIEA/DMF or DEPC/DIEA/DMF and prolonged reaction time (3-10 d) failed to give cyclopeptide **31**.

Since the cyclization of hexapeptides containing one Pro and several Aib residues remained difficult, the incorporation of two Pro units seemed reasonable.

As *cyclo*(Pro-Aib-Gly-Aib-Aib-Phe) **1** has been synthesized successfully, although in moderate cyclization yields (28-35%), an analogous peptide with Gly replaced by an additional Pro residue was prepared (*Scheme 7*). Thus, the reaction of **5** with **3** in THF (6 d) afforded tripeptide amide Fmoc-Pro-Aib-Aib-N(Me)Ph (**32**) in good yield. After removal of the Fmoc group of **32**, the HATU/HOAt-mediated coupling with Z-Phe-Pro-Aib-OH (**11**) gave hexapeptide Z-Phe-Pro-Aib-Pro-Aib-Aib-N(Me)Ph (**33**). The protecting groups were removed and the linear precursor subjected to cyclization, but no cyclic peptide **34** was formed after treatment with either PyAOP/HOAt or EDCI/HOAt for 3 d.

In summary, the cyclization of various hexapeptides containing Pro and three or four Aib residues has been investigated. In contrast to the general rule that Pro residues in linear peptides promote the formation of cyclization-prone conformations, our experimental data showed that the introduction of Pro into the peptide sequence containing Aib residues drastically hinders the formation of cyclic hexapeptides regardless of the position of Pro in the linear sequence. A total of 18 cyclization reactions were carried out in order to study the propensity of these hexapeptides to form cyclomonomers, but only three reactions were successful and resulted in moderate yields of cyclopeptides **1** or **2**. During cyclizations that fail to produce cyclomonomers, the formation of cyclic dimers or oligomers is commonly observed, but in the present study, the linear precursors either formed cyclomonomers in moderate yields or remained unaltered. One possible explanation for this observation might be the preference of rigid extended conformations when conformationally restricted proline along with

conformationally constrained Aib units are incorporated in short peptide sequences (*cf.* also [17]).

2.3. *Solid State Conformation of* Cyclo*(Phe-Pro-Aib-Gly-Aib-Aib-Phe)* **1**. Cyclic hexapeptides generally adopt conformations consisting of two β-turns, often referred to as the turn-extended-turn conformation [18]. A proline residue, in which the N-C(α) torsion angle ϕ is restricted to –60° (±20°) induces characteristic turn motifs within the cyclic peptides. As a consequence of this reduced conformational space, Pro strongly prefers the (*i* + 1)-position of most β-turns [19]. In Pro-Xaa sequences, the preference for a certain type of β-turn conformation depends on the structure of the Xaa residue, such as L/D configuration and side chain nature, as well as on the environment (solid or solution state, solvent polarity) [20].

We have carried out an X-ray crystallographic investigation of the molecular structure of **1** in order to study the effect that the Pro and Aib residues might have on the conformation of small cyclic peptides. Crystals were grown from the fraction containing **1** and some Et(*i*-Pr)$_2$NH$^+$PF$_6^-$ by slow evaporation of a solution in AcOEt/EtOH/hexane. The cyclic peptide **1** co-crystallized with Et(*i*-Pr)$_2$NH$^+$PF$_6^-$ and H_2O in a ratio 1:1:1. The ORTEP plot [21] of the molecular structure with the atom numbering scheme is presented in *Figure 1*. The relevant torsion angles for **1** are summarized in *Table 1*, and the H-bond parameters are shown in *Table 2*.

Fig. 1

The overall conformation of *cyclo*(Pro[1]-Aib[2]-Gly[3]-Aib[4]-Aib[5]-Phe[6]) (**1**) consists of two β-turns stabilized by a pair of intramolecular H-bonds involving the CO and NH groups of the Gly and Phe residues. The torsion angles ϕ and ψ of the Aib[4] and Aib[5] residues in the Gly[3]-Aib[4]-Aib[5]-Phe[6] segment show values close to those for a type I' β-turn (*Table 1*). The sequence Phe[6]-Pro[1]-Aib[2]-Gly[3] adopts a type III β-turn conformation with Pro[1] at the (*i* + 1)-position of the turn, as expected for this residue. All three Aib residues show torsion angles which are in good agreement with the expected values in the helical region of the conformational space. In addition, all the amide bonds have the *trans* conformation. Since structural information about the conformational preferences of Aib- and Pro-containing cyclic hexapeptides is scarce, our results cannot be compared with other data. To the best of our knowledge, the solution structure of *cyclo*(Pro-Leu-Aib)$_2$ is, so far, the only example of an Aib- and Pro-containing cyclic hexapeptide with a known conformation [22]. The NMR spectra and temperature coefficients of this cyclic peptide indicate an asymmetric conformation which contains both *cis* and *trans* Aib-Pro bonds. Therefore, the overall conformation consists of two β-turns of type II' and VI with Pro residues occupying the (*i* + 2)-positions of the turns.

Table 1

Table 2

We thank the analytical sections of our institute for spectra and analyses. Financial support from the *Swiss National Science Foundation* and *F. Hoffmann-La Roche AG*, Basel, is gratefully acknowledged.

Experimental Part

1. *General*. Solvents were purified by standard procedures. The used Aib-synthon was 2,2,*N*-trimethyl-*N*-phenyl-2*H*-azirin-3-amine (**3**) [23]. Thin-layer chromatography (TLC): *Merck* TLC aluminium sheets, silica gel *60* F_{254}. Column chromatography (CC): *Uetikon-Chemie* 'Chromatographiegel' *C-560*. M.p: *Büchi 510* apparatus; uncorrected. IR Spectra: *Perkin-Elmer-1600 FT-IR* spectrophotometer; in KBr; absorptions in cm^{-1}. ^{1}H- (300 MHz) and ^{13}C-NMR (75.5 MHz) Spectra: *Bruker ARX-300* instrument; in (D_6)DMSO at 300 K unless otherwise stated; δ in ppm, coupling constants *J* in Hz. MS: *Finnigan SSQ-700* or *MAT-90* instrument for CI; *Finnigan TSQ-700* triple quadrupole spectrometer for ESI; *m/z* (rel.%). Abbreviations: DEPBT: 3-(diethoxyphosphoryloxy)-1,2,3-benzotriazin-4(3*H*)-one, DEPC: diethylphosphorocyanidate, DIEA: *N*-ethyl-*N*,*N*-diisopropylamine, EDCI: *N*-ethyl-*N*'-(3-dimethylaminopropyl)carbodiimide, Fmoc: (fluoren-9-yl)methoxycarbonyl, HATU: *O*-(7-azabenzotriazol-1-yl)-*N*,*N*,*N*',*N*'-tetramethyluronium hexafluorophosphate, HOAt: 1-hydroxy-7-azabenzotriazole, HOBt: 1-hydroxybenzotriazole, PyAOP: (7-azabenzotriazol-1-yloxy)tris(pyrrolidino)phosphonium hexafluorophosphate, PyBOP: (1*H*-benzotriazol-1-yloxy)tris(pyrrolidino)phosphonium hexafluorophosphate, TBTU: 2-(1*H*-benzotriazol-1-yl)-1,1,3,3-tetramethyluronium tetrafluoroborate, Z: benzyloxycarbonyl.

General Procedure 1 (GP 1). To a soln. of an *N*-protected amino acid or peptide acid in abs. THF or MeCN was added the 2*H*-azirin-3-amine **3** (1.1 equiv.) in abs. THF, and the mixture was stirred at r.t. under N_2. After completion of the reaction (TLC), the solvent was evaporated i.v. and the residue purified by CC (SiO_2).

General Procedure 2 (GP 2). The peptide amide was dissolved in 3N HCl/MeCN 1:1 (*v*/*v*; 10 ml/mmol), and the soln. was stirred at r.t. overnight. The mixture was extracted with CH_2Cl_2 or AcOEt, dried (Na_2SO_4), and evaporated. The crude product was purified by CC or crystallization and dried u.h.v.

General Procedure 3 (GP 3). To a soln. of a Z-protected peptide in MeOH was added Pd/C (10%) and the mixture was hydrogenated overnight under atmospheric pressure using an H_2-filled balloon. The catalyst was removed by filtration through a pad of celite and the solvent evaporated. The residue was purified by chromatography on a short plug of SiO_2 using CH_2Cl_2/MeOH/NH_3(l) 12:1:0.1 as eluant.

General Procedure 4 (GP 4). To a soln. of an *N*-protected peptid acid in the given solvent were added the amino component (1.1 equiv.), coupling reagent (1.0-1.1 equiv.), additive (HOAt or HOBt, 1.0-1.1 equiv.) if indicated, and DIEA (2 equiv. without and 3 equiv. with hydrochloride salts present). The mixture was stirred at r.t. under N_2 until the starting material was consumed (TLC). The solvent was then evaporated, the residue was dissolved in AcOEt and washed with 5% aq. $KHSO_4$ soln., 5% aq. $NaHCO_3$ soln. and brine. The org. layer was dried ($MgSO_4$), concentrated, purified by CC and dried i.v.

General Procedure 5 (DEPBT-mediated Cyclization) (GP 5). Free linear hexapeptide was dissolved in abs. DMF (1.5 mM) under stirring. DEPBT (3-5 equiv.) and DIEA (1% *v*/*v*) were then added at r.t. and the solution was stirred for an additional 3 d. The solvent was evaporated under reduced pressure, and the crude cyclopeptide was purified by CC and prep. TLC.

General Procedure 6 (DEPC-mediated Cyclization) (GP 6). Free linear hexapeptide was dissolved in abs. DMF (1.0-1.5 mM) and the soln. was cooled to 0° in an ice bath. A soln. of 3-5 equiv. of DEPC in abs. DMF (1 ml) was added under stirring. Then, DIEA (1% *v*/*v*) was added slowly over a period of 15 min. The soln. was warmed to r.t. and stirred for an additional 6 d. The solvent was then evaporated under reduced pressure, the residue was taken up in AcOEt and washed with 5% aq. $KHSO_4$ soln., 5% aq. $NaHCO_3$ soln. and brine. The org. phase was dried ($MgSO_4$) and concentrated to give the crude cyclohexapeptide, which was purified by CC.

General Procedure 7 (PyAOP-mediated Cyclization) (GP 7). Free linear hexapeptide was dissolved in abs. DMF (0.5-1.0 mM) under stirring. PyAOP (3 or 5 equiv.), HOAt (3 or 5 equiv., 0.5 M soln. in DMF) and DIEA were then added at r.t. The soln. was stirred at r.t. for an additional 3 d. The solvent was then removed under reduced pressure, the residue dissolved in AcOEt and washed with 10% citric acid soln., 5% $NaHCO_3$ soln. and H_2O. The org. layer was dried (Na_2SO_4), filtered and concentrated to afford a yellow oil which was purified by CC.

2. *Preparation of Fmoc-Pro-Aib-Gly-Aib-Aib-Phe-OtBu* (**9**).

2.1. *Fmoc-Pro-Aib-N(Me)Ph* (**4**). According to the *GP 1* with Fmoc-Pro-OH (4.0 g, 11.85 mmol) in 50 ml abs. THF, **3** (2.272 g, 13.04 mmol) in 5 ml abs. THF, stirred for 60 h. CC (AcOEt/hexane 2:1) yielded 5.39 g (89%) of **4** as a white foam. IR: 3305*s*, 3063*m*, 2980*m*, 2948*m*, 2880*m*, 1703*s*, 1635*s*, 1594*s*, 1535*m*, 1493*s*, 1451*s*, 1417*s*, 1359*s*, 1289*m*, 1243*m*, 1192*m*, 1118*s*, 1090*s*, 988*m*, 760*s*, 741*s*, 705*m*.^{1}H-NMR: 8.09 (*s*, NH); 7.91-7.18 (*m*, 13 arom. H); 4.34-4.11 (*m*, C*H*C*H*$_2$O of Fmoc, CH(2) of Pro); 3.48-3.35 (*m*, CH$_2$(5) of Pro); 3.24 (*s*, MeN); 2.19-1.82 (*m*, CH$_2$(3) and CH$_2$(4) of Pro); 1.37, 1.35 (2*s*, 2 Me of Aib). ^{13}C-NMR: 172.1, 170.6 (2*s*, 2 CO); 154.0 (*s*, CO (urethane)); 145.6, 143.8, 143.6, 140.6 (4*s*, 5 arom. C); 128.5, 127.5, 127.3, 127.0, 126.2, 125.0, 120.0 (7*d*, 13 arom. CH); 66.4 (*t*, CH*C*H$_2$O of Fmoc); 59.4 (*d*, C(2) of Pro); 56.1 (*s*, C(2) of Aib); 46.5 (*d*, *C*HCH$_2$O of Fmoc); 46.3 (*t*, C(5) of Pro); 39.6 (*q*, MeN); 29.7 (*t*, C(3) of Pro); 26.0, 25.7 (2*q*, 2 Me of Aib); 23.7 (*t*, C(4) of Pro). ESI-MS (MeOH+NaI): 534 (100, [*M*+Na]$^{+}$). Anal. calc. for $C_{31}H_{33}N_3O_4$ (511.62): C 72.78, H 6.50, N 8.21; found: C 72.71, H 6.59, N 8.17.

2.2. *Fmoc-Pro-Aib-OH* (**5**). According to the *GP 2*, **4** (5.3 g, 10.35 mmol) in 3N HCl (MeCN/H$_2$O 1:1, 100 ml), stirred overnight. The product was extracted with AcOEt and purified by CC (AcOEt/MeOH 20:1) to give 3.22 g (74%) of pure **5**. White powder. M.p. 170.0-171.5°. IR: 3364*m*, 2987*m*, 2946*m*, 2895*m*, 2536*w*, 1706*s*, 1628*s*, 1528*s*, 1463*s*, 1451*s*, 1408*s*, 1356*m*, 1345*s*, 1328*s*, 1270*m*, 1252*m*, 1204*s*, 1175*s*, 1128*m*, 988*w*, 925*w*, 862*w*, 760*s*, 741*s*. ^{1}H-NMR: 8.21 (br.*s*, NH); 8.00-7.30 (*m*, 8 arom. H); 4.40-4.00 (*m*, C*H*C*H*$_2$O of Fmoc, CH(2) of Pro); 3.44-3.32 (*m*, CH$_2$(5) of Pro); 2.24-1.82 (*m*, CH$_2$(3) and CH$_2$(4) of Pro); 1.36, 1.33 (2*s*, 2 Me of Aib). ^{13}C-NMR: 175.3, 171.2 (2*s*, 2 CO); 153.8 (*s*, CO

(urethane)); 144.0, 143.7, 143.4, 140.6 (4*s*, 4 arom. C); 127.5, 127.0, 125.5, 125.2, 125.0, 120.0 (6*d*, 8 arom. CH); 66.7 (*t*, CH*C*H_2O of Fmoc); 59.2 (*d*, C(2) of Pro); 54.8 (*s*, C(2) of Aib); 47.0 (*t*, C(5) of Pro); 46.5 (*d*, *C*HCH_2O of Fmoc); 31.0 (*t*, C(3) of Pro); 25.1, 24.4 (2*q*, 2 Me of Aib); 22.7 (*t*, C(4) of Pro). ESI-MS (MeOH): 445 (48, $[M+Na]^+$), 423 (100, $[M+H]^+$). Anal. calc. for $C_{24}H_{26}N_2O_5$ (422.48): C 68.23, H 6.20, N 6.63; found: C 68.10, H 6.28, N 6.42.

2.3. *Fmoc-Pro-Aib-Gly-Aib-Aib-N(Me)Ph* (**7**). Z-Gly-Aib-Aib-N(Me)Ph (**6** [5], 0.54 g, 1.15 mmol) was *N*-deprotected following *GP 3* (H_2, 60 mg Pd/C, 6 ml MeOH, overnight). Purification by CC (CH_2Cl_2/MeOH 15:1) yielded 0.385 g (1.15 mmol, quant. yield) of H-Gly-Aib-Aib-N(Me)Ph as a white foam, which was used directly in the next step.

The coupling of Fmoc-Pro-Aib-OH (**5**, 0.442 g, 1.05 mmol) and the above-prepared H-Gly-Aib-Aib-N(Me)Ph (0.385 g, 1.15 mmol) was carried out using PyBOP (0.544 g, 1.05 mmol) and DIEA (0.27 g, 2.1 mmol) in 10 ml abs. CH_2Cl_2/MeCN (4:1) according to the *GP 4*. Purification by CC (AcOEt/MeOH 13:1, CH_2Cl_2/MeOH 15:1) yielded 0.536 g (69%) of **7**. White powder. M.p. 136-138°. IR: 3323*m*, 3064*w*, 2984*m*, 2940*m*, 1673*s*, 1594*m*, 1534*s*, 1494*m*, 1452*s*, 1425*m*, 1391*m*, 1361*m*, 1336*m*, 1281*m*, 1244*m*, 1195*m*, 1168*m*, 1125*m*, 1091*m*, 1017*w*, 991*w*, 760*m*, 741*m*, 708*m*. ^{1}H-NMR: 8.77, 8.04, 7.90, 7.88 (4*s*, 4 NH); 7.64-7.15 (*m*, 13 arom. H); 4.37-4.12 (*m*, C*HC*H_2O of Fmoc, CH(2) of Pro); 3.52-3.41 (*m*, CH_2 of Gly, CH_2(5) of Pro); 3.22 (*s*, MeN); 2.15-1.85 (*m*, CH_2(3) and CH_2(4) of Pro); 1.39, 1.36, 1.28, 1.21 (4*s*, 6 Me of 3 Aib). ^{13}C-NMR (CD_3OD): 178.1, 176.2, 175.5, 175.2, 171.6 (5*s*, 5 CO); 156.8 (*s*, CO(urethane)); 147.0, 145.0, 144.9, 142.5 (4*s*, 5 arom. C); 130.2, 128.9, 128.1, 126.2, 125.9, 121.0 (6*d*,

13 arom. CH); 69.0 (*t*, CH*C*H_2O of Fmoc); 61.5 (*d*, C(2) of Pro); 58.4, 58.2, 57.6 (3*s*, 3 C(2) of 3 Aib); 48.24 (*t*, C(5) of Pro); 48.20 (*d*, *C*HCH$_2$O of Fmoc); 45.4 (*t*, C(2) of Gly); 41.0 (*q*, MeN); 31.0 (*t*, C(3) of Pro); 25.6 (*t*, C(4) of Pro); 27.1, 26.5, 26.4, 26.1, 24.4, 24.0 (6*q*, 6 Me of 3 Aib). ESI-MS (MeOH/CH_2Cl_2): 784 (32, [*M*+K]$^+$), 632 (100, [*M*-N(Me)Ph]$^+$). Anal. calc. for $C_{41}H_{50}N_6O_7$•1/2H_2O (747.89): C 65.84, H 6.87, N 11.24; found: C 65.87, H 6.80, N 11.25.

2.4. *Fmoc-Pro-Aib-Gly-Aib-Aib-OH* (**8**). According to the *GP 2* with **7** (0.478 g, 0.65 mmol) in 6 ml 3N HCl (MeCN/H_2O 1:1), stirred overnight. The solvent was evaporated under reduced pressure and 5 ml of H_2O were added. The precipitated product was filtered, washed with H_2O and Et_2O and dried u.h.v. to give 0.38 g (91%) of pure **8**. White powder. M.p. 185.5-187.5°. IR: 3325*m*, 2983*m*, 2938*m*, 1662*s*, 1603*m*, 1525*s*, 1452*s*, 1362*m*, 1338*m*, 1289*m*, 1210*m*, 1133*m*, 1019*w*, 989*w*, 942*w*, 868*w*, 759*m*, 741*m*. ^{1}H-NMR (CD_3OD): 7.80-7.28 (*m*, 8 arom. H); 4.43-4.21 (*m*, C*HCH*$_2$O of Fmoc, CH(2) of Pro); 3.76-3.50 (*m*, CH_2 of Gly, CH_2(5) of Pro); 2.22-1.88 (*m*, CH_2(3) and CH_2(4) of Pro); 1.48, 1.43, 1.31, 1.19 (4*s*, 6 Me of 3 Aib). ^{13}C-NMR (CD_3OD): 181.3, 179.9, 176.1, 174.6, 169.7 (5*s*, 6 CO); 156.4 (*s*, CO(urethane)); 144.8, 143.1, 142.4 (3*s*, 4 arom. C); 128.9, 128.0, 127.7, 126.0, 125.7, 121.0, 120.9 (7*d*, 8 arom. CH); 70.2 (*t*, CH*C*H_2O of Fmoc); 60.7 (*d*, C(2) of Pro); 58.6, 58.3, 57.1 (3*s*, 3 C(2) of 3 Aib); 48.6 (*t*, C(5) of Pro); 47.4 (*d*, *C*HCH$_2$O of Fmoc); 44.7 (*t*, C(2) of Gly); 31.3 (*t*, C(3) of Pro); 25.7 (*t*, C(4) of Pro); 25.0, 24.3, 24.1, 22.9 (4*q*, 6 Me of 3 Aib). ESI-MS (MeOH+NaI): 672 (100, [*M*+Na]$^+$).

2.5. *Fmoc-Pro-Aib-Gly-Aib-Aib-Phe-OtBu* (**9**). According to the *GP 4*, coupling of **8** (0.338 g, 0.52 mmol) with HCl•H-Phe-O*t*Bu (0.147 g, 0.57 mmol),

using PyBOP (0.27 g, 0.52 mmol) and DIEA (0.2 g, 1.56 mmol) in 10 ml abs. CH_2Cl_2/MeCN (3:2). Reaction time: 20 h. The crude product was purified by CC (CH_2Cl_2/MeOH 20:1) to yield 0.358 g (81%) of **9**. White powder. M.p. 166.5-167.5°. IR: 3323*m*, 2981*m*, 2936*m*, 1671*s*, 1535*s*, 1453*s*, 1425*m*, 1363*m*, 1336*m*, 1283*m*, 1247*m*, 1160*s*, 1128*m*, 990*w*, 847*w*, 759*m*, 741*m*, 701*m*. ^{1}H-NMR (CD_3OD): 7.79-7.15 (*m*, 13 arom. H); 4.44-4.17 (*m*, C*H*C*H*$_2$O of Fmoc, CH(2) of Phe, CH(2) of Pro); 3.87-3.50 (*m*, CH_2 of Gly, CH_2(5) of Pro); 3.07-3.02 (*m*, CH_2 of Phe); 2.25-1.85 (*m*, CH_2(3) and CH_2(4) of Pro); 1.44, 1.42, 1.41, 1.35, 1.30, 1.15 (6*s*, 6 Me of 3 Aib, Me_3C). ^{13}C-NMR (CD_3OD): 177.9, 176.9, 175.8, 175.2, 172.2, 172.1 (6*s*, 6 CO); 156.8 (*s*, CO(urethane)); 145.0, 144.9, 142.6, 138.5 (4*s*, 5 arom. C); 130.4, 129.2, 128.9, 128.1, 127.5, 126.1, 125.9, 121.0 (8*d*, 13 arom. CH); 82.4 (*s*, Me_3C); 69.0 (*t*, CH*C*H$_2$O of Fmoc); 61.5, 56.5 (2*d*, C(2) of Pro, C(2) of Phe); 48.3 (*t*, C(5) of Pro); 48.2 (*d*, *C*HCH$_2$O of Fmoc); 45.3, 38.4 (2*t*, C(2) of Gly, C(3) of Phe); 31.0 (*t*, C(3) of Pro); 28.1 (*q*, *Me*$_3$C); 26.9, 26.8, 26.7 (3*q*, 6 Me of 3 Aib); 25.6 (*t*, C(4) of Pro). ESI-MS (MeOH+NaI): 876 (100, [*M*+Na]$^+$). Anal. calc. for $C_{47}H_{60}N_6O_9$•1/3H_2O (859.03): C 65.71, H 7.12, N 9.78; found: C 65.70, H 7.26, N 9.72.

3. *Preparation of Z-Phe-Pro-Aib-Gly-Aib-Aib-O*t*Bu* (**13**).

3.1. *Z-Phe-Pro-Aib-N(Me)Ph* (**10**). A soln. of Fmoc-Pro-Aib-N(Me)Ph (**4**, 2.479 g, 4.84 mmol) in 20 ml CH_2Cl_2/Et_2NH (1:1) was stirred for 3 h at r.t. The solvents were removed under reduced pressure and the crude *N*-deprotected peptide was purified by CC (CH_2Cl_2/MeOH 12:1) to give 1.21 g (86%) of H-Pro-Aib-N(Me)Ph, which was used directly in the next step.

The above-prepared dipeptide (1.21 g, 4.18 mmol) was coupled with Z-Phe-OH (1.138 g, 3.8 mmol) according to the *GP 4*, by using PyAOP (2.19 g, 4.2 mmol) and DIEA (1.36 g, 10.5 mmol) in 15 ml abs. CH_2Cl_2. Reaction time: 24 h. The crude product was purified by CC (AcOEt/MeOH 15:1, CH_2Cl_2/MeOH 35:1) to yield 1.94 g (89%) of pure **10** as a white foam. IR: 3416*m*, 3304*s*, 3061*m*, 3031*m*, 2981*m*, 2946*m*, 1715*s*, 1683*s*, 1643*s*, 1593*m*, 1531*s*, 1494*s*, 1453*s*, 1389*m*, 1361*m*, 1245*s*, 1211*m*, 1116*m*, 1089*m*, 1044*m*, 1027*m*, 1002*w*, 845*w*, 774*m*, 748*m*, 701*s*. ^{1}H-NMR: 8.06 (*s*, NH of Aib); 7.64 (*d*, *J* = 8.2, NH of Phe); 7.37-7.14 (*m*, 15 arom. H); 4.96 (*s*, PhC*H*$_2$O); 4.47-4.31 (*m*, CH(2) of Phe, CH(2) of Pro); 3.69-3.50 (*m*, CH_2(5) of Pro); 3.27 (*s*, MeN); 3.04-2.73 (*m*, CH_2 of Phe); 2.05-1.77 (*m*, CH_2(3) and CH_2(4) of Pro); 1.38, 1.37 (2*s*, 2 Me of Aib). ^{13}C-NMR: 172.1, 170.2, 170.1 (3*s*, 3 CO); 155.8 (*s*, CO(urethane)); 145.7, 137.8, 136.9 (3*s*, 3 arom. C); 129.1, 128.6, 128.1, 128.0, 127.6, 127.4, 127.3, 126.2 (8*d*, 15 arom. CH); 65.1 (*t*, PhC*H*$_2$O); 59.1 (*d*, C(2) of Pro); 56.0 (*s*, C(2) of Aib); 54.3 (*d*, C(2) of Phe); 46.6 (*t*, C(5) of Pro); 39.6 (*q*, MeN); 36.3 (*t*, C(3) of Phe); 28.7 (*t*, C(3) of Pro); 26.1, 25.4 (2*q*, 2 Me of Aib); 24.3 (*t*, C(4) of Pro). ESI-MS (MeOH+NaI): 593 (100, [*M*+Na]$^+$). Anal. calc. for $C_{33}H_{38}N_4O_5$•1/2H_2O (579.70): C 68.37, H 6.78, N 9.66; found: C 68.50, H 6.76, N 9.58.

3.2. *Z-Phe-Pro-Aib-OH* (**11**). According to the *GP 2*, **10** (0.68 g, 1.19 mmol) in 12 ml 3N HCl (MeCN/H_2O 1:1), stirred overnight. Purification by CC (CH_2Cl_2/MeOH 15:1) yielded 0.519 g (91%) of **11** as a white foam. IR: 3416*s*, 3063*m*, 3031*m*, 2982*m*, 1714*s*, 1640*s*, 1529*s*, 1454*s*, 1341*m*, 1247*s*, 1214*s*, 1162*s*, 1081*m*, 1051*m*, 1003*w*, 913*w*, 749*m*, 700*s*. ^{1}H-NMR: 7.94 (*s*, NH of Aib); 7.58 (*d*, *J* = 8.4, NH of Phe); 7.36-7.18 (*m*, 10 arom. H); 4.95 (br.*s*, PhC*H*$_2$O); 4.46-4.28

(*m*, CH(2) of Pro and CH(2) of Phe); 3.67-3.23 (*m*, CH_2(5) of Pro); 3.04-2.73 (*m*, CH_2(3) of Phe); 2.03-1.64 (*m*, CH_2(3) and CH_2(4) of Pro); 1.37, 1.35 (2*s*, 2 Me of Aib). ^{13}C-NMR: 175.9, 170.1, 170.0 (3*s*, 3 CO); 155.8 (*s*, CO(urethane)); 137.8, 136.9 (2*s*, 2 arom. C); 129.1, 128.1, 127.9, 127.5, 127.3, 126.1 (6*d*, 10 arom. CH); 65.1 (*t*, Ph*C*H_2O); 59.4 (*d*, C(2) of Pro); 55.0 (*s*, C(2) of Aib); 54.3 (*d*, C(2) of Phe); 46.6 (*t*, C(5) of Pro); 36.3 (*t*, C(3) of Phe); 28.5 (*t*, C(3) of Pro); 24.8, 24.2 (2*q*, 2 Me of Aib); 24.3 (*t*, C(4) of Pro). CI-MS (NH_3): 482 (27, $[M+H]^+$), 464 (100, $[M-OH]^+$). Anal. calc. for $C_{26}H_{31}N_3O_6 \bullet 1/2H_2O$ (490.55): C 63.66, H 6.57, N 8.56; found: C 63.57, H 6.32, N 8.37.

3.3. *Z-Phe-Pro-Aib-Gly-Aib-Aib-OtBu* (**13**). Z-Gly-Aib-Aib-OtBu (**12** [5], 0.425 g, 0.98 mmol) was *N*-deprotected (H_2, 50 mg Pd/C, 6 ml MeOH) according to the *GP 3*. 0.294 g (quant. yield) of H-Gly-Aib-Aib-O*t*Bu were obtained and used directly in the next step without further purification.

Coupling of the above deprotected tripeptide (0.294 g, 0.98 mmol) with **11** (0.427g, 0.89 mmol) was accomplished according to the *GP 4* by using HATU (0.342 g, 0.9 mmol), HOAt (0.9 mmol, 1.8 ml 0.5M DMF-solution) and DIEA (0.232 g, 1.8 mmol) in 10 ml abs. CH_2Cl_2. Reaction time: 20 h. The crude material was purified by CC (CH_2Cl_2/MeOH 15:1) to give 0.495 g (73%) of pure **13** as a white foam. IR: 3328*s*, 3063*w*, 3031*w*, 2982*m*, 2937*m*, 1670*s*, 1529*s*, 1454*s*, 1386*m*, 1367*m*, 1310*m*, 1252*m*, 1149*s*, 1083*w*, 1044*m*, 919*w*, 850*s*, 751*m*, 699*m*. ^{1}H-NMR: 8.90 (*s*, NH of Aib); 8.27 (*t-like*, NH of Gly); 7.74 (*d*, *J* = 8.5, NH of Phe); 7.46 (*s*, NH of Aib); 7.35-7.21 (*m*, 10 arom. H); 7.13 (*s*, NH of Aib); 4.93-4.92 (*m*, PhC*H*$_2$O); 4.45-4.37 (*m*, CH(2) of Pro and CH(2) of Phe); 3.81-3.50 (*m*, CH_2(5) of Pro and CH_2 of Gly); 2.89-2.66 (*m*, CH_2 of Phe); 2.18-1.80 (*m*, CH_2(3)

and CH_2(4) of Pro); 1.43, 1.37, 1.32, 1.30, 1.29, 1.26 (6*s*, 6 Me of 3 Aib, Me_3C). ^{13}C-NMR: 175.7, 173.0, 172.8, 171.0, 168.5 (5*s*, 6 CO); 155.9 (*s*, CO(urethane)); 137.5, 136.8 (2*s*, 2 arom. C); 129.1, 128.2, 128.1, 127.7, 127.5, 126.4 (6*d*, 10 arom. CH); 78.9 (*s*, Me_3C); 65.3 (*t*, PhCH_2O); 60.0 (*d*, C(2) of Pro); 56.0, 55.7, 55.2 (3*s*, 3 C(2) of 3 Aib); 54.5 (*d*, C(2) of Phe); 47.0 (*t*, C(5) of Pro); 44.4 (*t*, C(2) of Gly); 36.4 (*t*, C(3) of Phe); 28.3 (*t*, C(3) of Pro); 27.4 (*q*, Me_3C); 25.0 (*t*, C(4) of Pro); 26.6, 25.9, 25.1, 23.8, 23.6, 23.4 (6*q*, 6 Me of 3 Aib). ESI-MS (MeOH+NaI): 788 (100, $[M+Na]^+$).

4. *Synthesis of* Cyclo*(Pro-Aib-Gly-Aib-Aib-Phe)* (**1**).

4.1. Cyclo*(Pro-Aib-Gly-Aib-Aib-Phe)* (**1**) *from* **9**. To a soln. of **9** (0.318 g, 0.37 mmol) in 4 ml MeCN was added 1.2 ml Et_2NH at r.t. and the mixture was stirred for 3 h. The solvents were then removed under reduced pressure and the residue purified by CC (CH_2Cl_2/MeOH 15:1) to give 0.227 g (96%) H-Pro-Aib-Gly-Aib-Aib-Phe-O*t*Bu as a white foam. This *N*-deprotected hexapeptide (0.227 g, 0.35 mmol) was dissolved in 10 ml CH_2Cl_2 and TFA (10 ml) was then added. The mixture was stirred for 5 h at r.t. Excess TFA was evaporated under reduced pressure and the remaining TFA was removed by co-evaporation with Et_2O (3x 5 ml). The residue was triturated with Et_2O, the soln. decanted, and the operation was repeated twice to afford, after drying i.v., 0.257 g (0.35 mmol, quant. yield) of the deprotected hexapeptide as its TFA salt. 0.117 g (0.15 mmol) of this material was dissolved in 100 ml of abs. THF and subjected to cyclization with DEPBT (0.14 g, 0.46 mmol) and DIEA (1 ml) according to the *GP 5*. Reaction time: 3 d. Purification by CC (CH_2Cl_2/MeOH 12:1, 2x) and preparative TLC

(CH_2Cl_2/MeOH 10:1) afforded 30 mg (35%) of pure **1** as a white powder. M.p. >280° (dec.) (for data of **1** see 4.2).

4.2. Cyclo*(Pro-Aib-Gly-Aib-Aib-Phe)* (**1**) *from* **13**. The N-terminus of **13** (0.4 g, 0.52 mmol) was deprotected according to the *GP 3* (H_2, 45 mg Pd/C, 10 ml MeOH) to give 0.328 g (quant. yield) of H-Phe-Pro-Aib-Gly-Aib-Aib-O*t*Bu as a pale yellow foam. This material was dissolved in 30 ml CH_2Cl_2/TFA (1:1) and stirred for 4 h at r.t. Excess TFA was removed as described in 4.1 and 0.404 g (0.52 mmol, quant. yield) of the deprotected linear precursor was obtained as its TFA salt, which was used in the cyclization reactions without further purification.

DEPC-mediated cyclization: A soln. of 0.135 g (0.17 mmol) of the above prepared deprotected hexapeptide in 175 ml of abs. DMF was subjected to cyclization with DEPC (0.141 g, 0.86 mmol) and DIEA (1.75 ml) according to the *GP 6*. Reaction time: 6 d. Purification by CC (CH_2Cl_2/MeOH 13:1, then CH_2Cl_2/MeOH 17:1) afforded 33 mg (34%) of pure **1** as a white foam.

PyAOP-mediated cyclization: A soln. of 0.135 g (0.17 mmol) of the linear precursor in 220 ml of abs. DMF was subjected to cyclization using PyAOP (0.361 g, 0.69 mmol) and DIEA (2.2 ml) according to the *GP 7*. Reaction time: 24 h. Purification by CC (AcOEt/MeOH 10:1, then CH_2Cl_2/MeOH 15:1) afforded 27 mg (28%) pure **1** as a white foam. In addition, 15 mg of **1** contaminated with some Et(*i*-Pr)$_2$NH$^+$PF$_6^-$ was obtained, but could not be purified by CC. Crystals suitable for an X-ray crystal structure analysis were obtained from this mixture by crystallization from AcOEt/EtOH/hexane.

DEPBT-mediated cyclization: A soln. of 0.134 g (0.17 mmol) of the linear hexapeptide in 175 ml of abs. DMF was subjected to cyclization with DEPBT

(0.26 g, 0.86 mmol) and DIEA (1.75 ml) according to the *GP 5*. No cyclic product was obtained after 3 d reaction time.

Data of *cyclo*(Pro[1]-Aib[2]-Gly[3]-Aib[4]-Aib[5]-Phe[6]) (**1**): IR: 3317*s*, 3061*w*, 1985*m*, 2938*m*, 1665*s*, 1536*s*, 1444*m*, 1385*m*, 1363*m*, 1285*m*, 1216*m*, 1191*m*, 1101*w*, 1019*w*, 923*w*, 875*w*, 825*w*, 747*w*, 701*w*. ^{1}H-NMR: 9.30 (*s*, NH of Aib[2]); 8.46 (*t*-like, NH of Gly[3]); 7.90 (*s*, NH of Aib[4]); 7.42 (*d*, *J* = 8.4, NH of Phe[6]); 7.40-7.31 (*m*, 2 arom. H of Phe[6]); 7.25-7.12 (*m*, NH of Aib[5], 3 arom. H of Phe[6]); 4.58-4.55 (*m*, CH(2) of Phe[6]); 4.36-4.33 (*m*, CH(2) of Pro[1]); 3.86-3.79 (*m*, 1H of CH_2 of Gly[3], 1H of CH_2(5) of Pro[1]); 3.73-3.66 (*m*, 1H of CH_2 of Gly[3], 1H of CH_2(5) of Pro[1]); 2.86-2.67 (*m*, CH_2 of Phe[6]); 2.20-2.11 (*m*, 1H of CH_2(3) and 1H of CH_2(4) of Pro[1]); 1.98-1.93 (*m*, 1H of CH_2(4) of Pro[1]); 1.89-1.82 (*m*, 1H of CH_2(3) of Pro[1]); 1.46, 1.39 (2*s*, 2 Me of Aib[2]); 1.35 (*s*, Me of Aib[4]); 1.31, 1.30 (2*s*, 2 Me of Aib[5]); 1.18 (*s*, Me of Aib[4]). ^{13}C-NMR: 176.3 (*s*, CO of Aib[2]); 174.4 (*s*, CO of Aib[5]); 173.8 (*s*, CO of Pro[1]); 172.5 (*s*, CO of Aib[4]); 171.7 (*s*, CO of Phe[6]); 170.1 (*s*, CO of Gly[3]); 137.3 (*s*, 1 arom. C of Phe[6]); 129.3, 128.1, 126.2 (3*d*, 5 arom. CH of Phe[6]); 59.7 (*d*, C(2) of Pro[1]); 56.1 (*s*, C(2) of Aib[4]); 56.0 (*s*, C(2) of Aib[2]); 55.8 (*s*, C(2) of Aib[5]); 53.1 (*d*, C(2) of Phe[6]); 47.3 (*t*, C(5) of Pro[1]); 45.1 (*t*, C(2) of Gly[3]); 36.9 (*t*, C(3) of Phe[6]); 28.4 (*t*, C(3) of Pro[1]); 27.8 (*q*, Me of Aib[5]); 27.4 (*q*, Me of Aib[4]); 26.5 (*q*, Me of Aib[2]); 24.9 (*t*, C(4) of Pro[1]); 23.1 (*q*, Me of Aib[2]); 22.4 (*q*, Me of Aib[4]); 22.3 (*q*, Me of Aib[5]). ESI-MS (MeOH + NaI): 579 (100, $[M+Na]^+$).

5. *Preparation of Z-Gly-Aib-Aib-Phe-Pro-Aib-N(Me)Ph* (**14**).

5.1. *Synthesis of* **14**. Deprotection of the N-terminus of **10** (0.728 g, 1.27 mmol) was performed according to the *GP 3* (H_2, 80 mg Pd/C, 10 ml MeOH).

Purification by CC (CH_2Cl_2/MeOH 10:1) yielded 0.519 g (93%) of H-Phe-Pro-Aib-N(Me)Ph, which was used directly in the next step.

The above obtained deprotected tripeptide (0.519 g, 1.19 mmol) was dissolved in 10 ml of abs. CH_2Cl_2 and coupled with Z-Gly-Aib-Aib-OH [5] (0.41 g, 1.08 mmol) using PyAOP (0.573 g, 1.1 mmol), HOAt (0.15 g, 1.1 mmol, 2.2 ml 0.5M DMF-soln.), and DIEA (0.284 g, 2.2 mmol) according to the *GP 4*. Reaction time: 20 h. Purification of the crude product by CC (CH_2Cl_2/MeOH 15:1) yielded 0.78 g (90%) of pure **14** as a white foam. IR: 3322*s*, 3061*m*, 3031*m*, 2984*m*, 2937*m*, 1650*s*, 1593*m*, 1522*s*, 1496*s*, 1454*s*, 1385*m*, 1362*m*, 1237*s*, 1172*m*, 1116*m*, 1090*m*, 1048*m*, 1001*w*, 922*w*, 774*w*, 742*m*, 702*m*. ^{1}H-NMR: 8.09, 7.94 (2*s*, 2 NH); 7.53-7.12 (*m*, 15 arom. H, 3 NH); 5.02 (*s*, PhC*H*$_2$O); 4.65-4.60, 4.25-4.23 (2*m*, CH(2) of Phe, CH(2) of Pro); 3.68-3.41 (*m*, CH_2 of Gly and CH_2(5) of Pro); 3.23 (*s*, MeN); 3.06-2.81 (*m*, CH_2 of Phe); 1.98-1.78 (*m*, CH_2(3) and CH_2(4) of Pro); 1.35, 1.30, 1.27, 1.23 (4*s*, 6 Me of Aib). ^{13}C-NMR: 173.8, 172.8, 172.2, 170.4, 169.3, 169.1 (6*s*, 6 CO); 156.6 (*s*, CO (urethane)); 145.7, 137.8, 136.9 (3*s*, 3 arom. C); 129.3, 128.6, 128.2, 127.9, 127.7, 127.5, 127.2, 126.8, 126.2, 126.0 (10*d*, 15 arom. CH); 66.5 (*t*, PhC*H*$_2$O); 59.3 (*d*, C(2) of Pro); 56.0, 55.8 (2*s*, 3 C(2) of 3 Aib); 52.3 (*d*, C(2) of Phe); 46.4, 43.9 (2*t*, C(2) of Gly and C(5) of Pro); 39.5 (*q*, MeN); 36.6 (*t*, C(3) of Phe); 28.6 (*t*, C(3) of Pro); 26.1, 25.5, 24.7, 24.3 (4*q*, 6 Me of 3 Aib); 24.2 (*t*, C(4) of Pro). ESI-MS (MeOH+NaI): 820 (100, [*M*+Na]$^+$). Anal. calc. for $C_{43}H_{55}N_7O_8$•1/2H_2O (806.96): C 64.00, H 6.99, N 12.15; found: C 63.86, H 6.96, N 12.07.

5.2. *Attempted Cyclization of* **14**. According to the *GP 2*, a soln. of **14** (0.68 g, 0.85 mmol) in 10 ml of 3N HCl (MeCN/H_2O 1:1) was stirred for 15 h.

Then, the MeCN was evaporated, 2N HCl (5 ml) was added and the product was extracted with CH_2Cl_2, dried (Na_2SO_4), concentrated and dried i.v.: 0.54 g (89%) of peptide acid were obtained as a white foam. This acid (0.464 g, 0.65 mmol) was hydrogenated according to the *GP 3* (H_2, 50 mg Pd/C, 8 ml MeOH) to give 0.358 g (95%) of H-Gly-Aib-Aib-Phe-Pro-Aib-OH as a pale yellow foam, which was used in the cyclization reactions without further purification.

DEPC-mediated cyclization: 90 mg (0.16 mmol) of the linear hexapeptide was dissolved in abs. DMF (160 ml) and subjected to cyclization with DEPC (0.128 g, 0.78 mmol) and DIEA (1.6 ml) according to the *GP 6*. No cyclopeptide **1** was formed after 3 d reaction time.

DEPBT-mediated cyclization: Following the *GP 5*, with 90 mg (0.16 mmol) of the linear peptide dissolved in abs. DMF (100 ml), DEPBT (0.141 g, 0.47 mmol) and DIEA (1 ml). No cyclic product was obtained after 3 d reaction time.

PyAOP-mediated cyclization: According to the *GP 7*, with 90 mg (0.16 mmol) of the linear precursor in abs. DMF (160 ml) and PyAOP (0.328 g, 0.63 mmol), HOAt (0.63 mmol, 1.4 ml 0.5M DMF-soln.) and DIEA (1.6 ml). No cyclic product was obtained after 3 d reaction time.

HATU-mediated cyclization: The linear hexapeptide (74 mg, 0.13 mmol) was dissolved in abs. DMF (130 ml) and cooled to 0° in an ice bath. HATU (54 mg, 0.14 mmol), HOAt (0.14 mmol, 0.3 ml 0.5M DMF-soln.), and collidine (0.157 g, 13 mmol) were then added under stirring. The mixture was allowed to warm to r.t. over a period of 2 h, and was stirred for a further 3 d. No formation of cyclopeptide **1** was observed.

6. *Synthesis of* Cyclo*(Gly-Aib-Pro-Gly-Aib-Aib)* (**2**).

6.1. *Fmoc-Pro-Gly-Aib-Aib-N(Me)Ph* (**15**). Deprotection of the N-terminus of **6** (0.4 g, 0.85 mmol) was accomplished by following the *GP 3* (H_2, 45 mg Pd/C, 10 ml MeOH). The obtained H-Gly-Aib-Aib-N(Me)Ph (0.285 g, quant. yield) was used directly in the next step without further purification.

Coupling of Fmoc-Pro-OH (0.262 g, 0.78 mmol) with the above prepared *N*-deprotected peptide (0.285 g, 0.85 mmol) was performed using PyAOP (0.47 g, 0.9 mmol) and DIEA (0.233 g, 1.8 mmol) in abs. CH_2Cl_2 (10 ml) according to the *GP 4*. Reaction time: 24 h. Purification by CC (AcOEt/MeOH 15:1, CH_2Cl_2/MeOH 20:1) yielded 0.47 g (93%) of pure **15** as a white foam. IR: 3748*m*, 3671*m*, 3647*m*, 3419*s*, 3064*m*, 2983*m*, 2934*m*, 1675*s*, 1594*m*, 1524*s*, 1494*s*, 1452*s*, 1423*s*, 1391*m*, 1360*s*, 1339*m*, 1195*m*, 1168*m*, 1124*m*, 1092*m*, 1040*w*, 1022*w*, 989*w*, 759*m*, 740*m*, 706*m*. ^{1}H-NMR (CD_3OD): 7.81-7.18 (*m*, 13 arom. H); 4.47-4.20 (*m*, C*H*CH_2O of Fmoc and CH(2) of Pro); 3.74-3.47 (*m*, CH_2 of Gly and CH_2(5) of Pro); 3.22 (*s*, MeN); 2.30-1.80 (*m*, CH_2(3) and CH_2(4) of Pro); 1.46, 1.45, 1.41, 1.39 (4*s*, 4 Me of 2 Aib). ^{13}C-NMR (CD_3OD): 175.9, 175.3, 171.0 (3*s*, 4 CO); 157.0 (*s*, CO(urethane)); 145.1, 142.5 (2*s*, 5 arom. C); 130.2, 128.8, 128.4, 128.1, 125.9, 120.9 (6*d*, 13 arom. CH); 68.8 (*t*, CH*C*H_2O of Fmoc); 62.1 (*d*, C(2) of Pro); 58.6, 58.2 (2*s*, 2 C(2) of 2 Aib); 48.3 (*d*, *C*HCH_2O of Fmoc); 48.1, 44.6 (2*t*, C(5) of Pro and C(2) of Gly); 41.2 (*q*, MeN); 31.1 (*t*, C(3) of Pro); 26.2, 25.9, 25.3 (3*q*, 4 Me of 2 Aib); 25.5 (*t*, C(4) of Pro). ESI-MS (MeOH+NaI): 676 (100, [*M*+Na]$^+$), 534 (70, [*M*-N(Me)Ph]$^+$). Anal. calc. for $C_{37}H_{43}N_5O_6$•1/3H_2O (659.78): C 67.36, H 6.67, N 10.61; found: C 67.27, H 6.90, N 10.61.

6.2. *Z-Gly-Aib-Pro-Gly-Aib-Aib-N(Me)Ph* (**16**). To a soln. of **15** (0.47 g, 0.72 mmol) in CH_2Cl_2 (7 ml), Et_2NH (5 ml) was added under stirring. After 5 h at

r.t. the solvents were removed under reduced pressure and the residue was purified by CC (CH_2Cl_2/MeOH 10:1) to give 0.253 g (82%) of H-Pro-Gly-Aib-Aib-N(Me)Ph, which was used directly in the next step.

Coupling of Z-Gly-Aib-OH (0.157 g, 0.53 mmol) with the above-obtained tripeptide (0.253 g, 0.58 mmol) in abs. CH_2Cl_2 (10 ml) was carried out using PyAOP (0.313 g, 0.6 mmol) and DIEA (0.156 g, 1.2 mmol) according to the *GP 4*. Reaction time: 24 h. The crude product was purified by CC (CH_2Cl_2/MeOH 15:1) to give 0.33 g (87%) of **16** as a white foam. IR: 3745*m*, 3418*s*, 3062*m*, 2985*m*, 2936*m*, 1659*s*, 1594*m*, 1529*s*, 1471*m*, 1455*m*, 1395*s*, 1364*m*, 1244*s*, 1171*m*, 1115*m*, 1092*m*, 1053*m*, 769*w*, 741*w*, 706*w*. ^{1}H-NMR (CD_3OD): 7.55-7.24 (*m*, 10 arom. H); 5.13-5.03 (*m*, PhC*H*$_2$O); 4.39-4.34 (*m*, CH(2) of Pro); 3.92-3.43 (*m*, 2 CH_2 of 2 Gly and CH_2(5) of Pro); 3.34 (*s*, MeN); 2.27-1.74 (*m*, CH_2(3) and CH_2(4) of Pro); 1.52, 1.49, 1.48, 1.47, 1.45 (5*s*, 6 Me of 3 Aib). ^{13}C-NMR (CD_3OD): 176.2, 175.6, 175.5, 174.9, 172.2, 171.3 (6*s*, 6 CO); 158.5 (*s*, CO(urethane)); 146.8, 138.0 (2*s*, 2 arom. C); 130.2, 129.4, 129.1, 128.8, 128.3, 128.1 (6*d*, 10 arom. CH); 67.7 (*t*, PhC*H*$_2$O); 64.5 (*d*, C(2) of Pro); 58.6, 58.3, 57.7 (3*s*, 3 C(2) of 3 Aib); 49.7 (*t*, C(5) of Pro); 44.5, 44.3 (2*t*, 2 C(2) of 2 Gly); 41.1 (*q*, MeN); 29.5, 26.8 (2*t*, C(3) and C(4) of Pro); 26.5, 26.4, 25.0, 23.9 (4*q*, 6 Me of 3 Aib). ESI-MS (MeOH+NaI): 730 (100, $[M+Na]^+$). Anal. calc. for $C_{36}H_{49}N_7O_8 \bullet 1/2H_2O$ (716.83): C 60.32, H 7.03, N 13.68; found: C 60.27, H 6.99, N 13.61.

6.3. *Z-Gly-Aib-Pro-Gly-Aib-Aib-OH* (**17**). According to the *GP 2*, **16** (0.25 g, 0.35 mmol) in 4 ml of 3N HCl (MeCN/H_2O 1:1) was stirred overnight. The product was extracted with CH_2Cl_2, dried (Na_2SO_4) and evaporated: 0.177 g

(82%) of pure **17** were obtained as a white foam. IR: 3304*s*, 3061*m*, 2986*m*, 2940*m*, 1719*s*, 1657*s*, 1534*s*, 1470*m*, 1455*m*, 1410*m*, 1365*m*, 1243*s*, 1165*s*, 1051*m*, 910*w*, 777*w*, 745*w*, 697*m*. ^{1}H-NMR (CD_3OD): 8.61, 7.99, 7.62 (3br.*s*, 3 NH); 7.36-7.27 (*m*, 5 arom. H); 5.14-5.03 (*m*, PhC*H*$_2$O); 4.40-4.35 (*m*, CH(2) of Pro); 3.92-3.30 (*m*, CH_2(5) of Pro and 2 CH_2 of 2 Gly); 2.26-1.73 (*m*, CH_2(3) and CH_2(4) of Pro); 1.50, 1.475, 1.468, 1.46, 1.44 (5*s*, 6 Me of 3 Aib). ^{13}C-NMR (CD_3OD): 177.9, 176.0, 175.5, 174.9, 172.3, 171.3 (6*s*, 6 CO); 159.0 (*s*, CO(urethane)); 138.1 (*s*, 2 arom. C); 129.4, 129.1, 128.8 (3*d*, 5 arom. CH); 67.7 (*t*, Ph*C*H$_2$O); 64.4 (*d*, C(2) of Pro); 58.1, 57.8, 57.1 (3*s*, 3 C(2) of 3 Aib); 49.7 (*t*, C(5) of Pro); 44.3, 44.2 (2*t*, 2 C(2) of 2 Gly); 29.5, 26.8 (2*t*, C(3) and C(4) of Pro); 26.4, 25.9, 25.3, 25.1, 25.0, 23.9 (6*q*, 6 Me of 3 Aib). ESI-MS (MeOH+NaI): 641 (100, [*M*+Na]$^+$). Anal. calc. for $C_{29}H_{42}N_6O_9$ (618.69): C 56.29, H 6.84, N 13.58; found: C 55.98, H 6.68, N 13.46.

6.4. Cyclo*(Gly*^{1}*-Aib*^{2}*-Pro*^{3}*-Gly*^{4}*-Aib*^{5}*-Aib*^{6}*)* (**2**). Peptide acid **17** (0.162 g, 0.26 mmol) was hydrogenated according to the *GP 3* (H_2; 25 mg Pd/C, 6 ml MeOH) to give 0.125 g (0.26 mmol, quant. yield) of the deprotected hexapeptide as a pale yellow foam, which was used in the cyclization step without further purification. According to the *GP 6*, this material (0.125 g, 0.26 mmol) in abs. DMF (170 ml) was treated with DEPC (0.168 g, 1.03 mmol) and DIEA (1.7 ml). Reaction time: 7 d. During the evaporation of DMF, an extremely insoluble white precipitate formed. This precluded its purification by CC. The precipitate was collected by filtration and washed with H_2O, MeOH and CH_2Cl_2 in order to remove excess DEPC and some formed byproducts. Recrystallization from hot H_2O/MeOH/EtOH yielded 43 mg (36%) of pure **2** as a white powder. M.p. >320°

(dec.). IR: 3487*m*, 3365*m*, 3324*s*, 3283*s*, 3044*w*, 2990*w*, 2937*w*, 1683*s*, 1650*s*, 1598*s*, 1545*s*, 1470*m*, 1418*m*, 1385*m*, 1366*m*, 1337*w*, 1252*m*, 1194*w*, 1178*w*, 1021*w*, 944*w*, 919*w*, 811*w*, 705*w*. ^{1}H-NMR ((D_6)DMSO + 3 drops of TFA): 8.78 (*s*, NH of Aib^2); 8.31 (*s*, NH of Aib^5); 7.49 (*s*, NH of Aib^6); 7.42 (br.*d*, *J* = 9.4, NH of Gly^4); 7.36 (br.*d*, *J* = 9.1, NH of Gly^1); 4.67-4.61 (*m*, CH(2) of Pro^3); 4.30 (*dd*, *J* = 9.8, 17.5, 1H of CH_2 of Gly^4); 4.22 (*dd*, *J* = 9.2, 17.8, 1H of CH_2 of Gly^1); 3.73-3.67 (*m*, 1H of CH_2(5) of Pro^3); 3.53 (br.*d*, *J* = 17.3, 1H of CH_2 of Gly^1); 3.48-3.35 (*m*, 1H of CH_2 of Gly^4 and 1H of CH_2(5) of Pro^3); 1.99-1.92 (*m*, 1H of CH_2(3) of Pro^3); 1.88-1.79 (*m*, 1H of CH_2(3) and 1H of CH_2(4) of Pro^3); 1.71-1.62 (*m*, 1H of CH_2(4) of Pro^3); 1.45 (*s*, Me of Aib^6); 1.36, 1.34 (2*s*, 2 Me of Aib^2); 1.31 (*s*, Me of Aib^5); 1.27 (br.*s*, Me of Aib^5 and Me of Aib^6). ^{13}C-NMR ((D_6)DMSO + 3 drops of TFA): 174.1 (*s*, CO of Aib^6); 173.5 (*s*, CO of Aib^5); 171.8 (*s*, CO of Aib^2); 171.1 (*s*, CO of Pro^3); 169.7 (*s*, CO of Gly^4); 169.1 (*s*, CO of Gly^1); 61.1 (*d*, C(2) of Pro^3); 56.9 (*s*, C(2) of Aib^6); 56.3 (*s*, C(2) of Aib^5); 55.9 (*s*, C(2) of Aib^2); 46.8 (*t*, C(5) of Pro^3); 39.7 (*t*, C(2) of Gly^4); 39.2 (*t*, C(2) of Gly^1); 28.5 (*q*, Me of Aib^6); 27.7 (*t*, C(3) of Pro^3); 26.4 (*q*, Me of Aib^5); 26.0 (*q*, Me of Aib^2); 24.3 (*t*, C(4) of Pro^3); 23.4 (*q*, Me of Aib^2); 23.25 (*q*, Me of Aib^5); 23.1 (*q*, Me of Aib^6). ESI-MS (MeOH + NaI): 489 (100, $[M+Na]^+$).

7. *Synthesis of Z-Gly-Aib-Pro-Aib-Phe-Aib-O*t*Bu* (**21**).

7.1. *Z-Gly-Aib-Pro-Aib-N(Me)Ph* (**18**). A soln. of **4** (0.71 g, 1.39 mmol) in CH_2Cl_2 (10 ml) and Et_2NH (5 ml) was stirred for 3 h at r.t. The solvents were then removed under reduced pressure and the crude *N*-deprotected peptide was purified by CC (CH_2Cl_2/MeOH/NH_3(l) 10:1:0.1) to give 0.322 g (80%) H-Pro-Aib-N(Me)Ph, which was used directly in the next step.

The coupling of this dipeptide (0.23 g, 0.79 mmol) with Z-Gly-Aib-OH (0.213 g, 0.72 mmol) in abs. CH_2Cl_2 (6 ml) and abs. MeCN (2 ml) was accomplished using PyAOP (0.417 g, 0.8 mmol) and DIEA (0.206 g, 1.6 mmol) according to the *GP 4*. Reaction time: 18 h. Purification by CC (CH_2Cl_2/MeOH 20:1, 2x) yielded 0.385 g (94%) **18** as a white foam. IR: 3423*s*, 3299*s*, 3062*m*, 3037*m*, 2985*m*, 2939*m*, 2878*w*, 1722*s*, 1647*s*, 1594*m*, 1534*s*, 1495*s*, 1470*m*, 1454*m*, 1399*s*, 1363*s*, 1242*s*, 1212*m*, 1168*m*, 1115*m*, 1090*m*, 1051*m*, 927*w*, 769*w*, 741*w*. ^{1}H-NMR: 8.53, 7.58 (2*s*, 2 NH); 7.49 (*t*, *J* = 6.1, NH of Gly); 7.39-7.15 (*m*, 10 arom. H); 5.10-4.96 (*m*, PhC*H*$_2$O); 4.33-4.28 (*m*, CH(2) of Pro); 3.84, 3.78 (2*d*, *J* = 6.3, CH_2 of Gly); 3.67-3.45 (*m*, CH_2(5) of Pro); 3.26 (*s*, MeN); 2.09-1.60 (*m*, CH_2(3) and CH_2(4) of Pro); 1.40, 1.37, 1.36, 1.32 (4*s*, 4 Me of 2 Aib). ^{13}C-NMR: 172.5, 171.4, 170.8, 169.1 (4*s*, 4 CO); 156.4 (*s*, CO(urethane)); 146.1, 136.8 (2*s*, 2 arom. C); 128.5, 128.2, 127.7, 127.5, 126.7, 125.7 (6*d*, 10 arom. CH); 65.3 (*t*, PhC*H*$_2$O); 61.5 (*d*, C(2) of Pro); 55.7, 55.6 (2*s*, 2 C(2) of 2 Aib); 47.4, 42.6 (2*t*, C(5) of Pro and C(2) of Gly); 39.1 (*q*, MeN); 28.2 (*t*, C(3) of Pro); 25.7, 25.5, 25.2, 23.8 (4*q*, 4 Me of 2 Aib); 25.1 (*t*, C(4) of Pro). ESI-MS (MeOH+NaI): 588 (100, [*M*+Na]$^+$). Anal. calc. for $C_{30}H_{39}N_5O_6 \bullet 1/3H_2O$ (571.67): C 63.03, H 6.99, N 12.25; found: C 63.04, H 6.91, N 12.28.

7.2. *Z-Gly-Aib-Pro-Aib-OH* (**19**). According to the *GP 2*, with 0.47 g (0.83 mmol) of **18** in 3N HCl (MeCN/H_2O 1:1) (10 ml), stirred for 24 h. The product was extracted from the mixture with CH_2Cl_2, dried (Na_2SO_4), and concentrated. Purification by CC (CH_2Cl_2/MeOH 11:1) yielded 0.26 g (66%) of **19** as a white foam. IR: 3415*s*, 3311*s*, 3063*m*, 2986*s*, 2942*s*, 1723*s*, 1657*s*, 1536*s*, 1470*m*, 1455*m*, 1406*s*, 1365*s*, 1343*m*, 1245*s*, 1161*s*, 1051*m*, 977*w*, 931*w*, 778*w*, 741*m*,

698*m*. ^{1}H-NMR (CD_3OD): 7.97, 7.78 (2*s*, 2 NH); 7.34-7.28 (*m*, 5 arom. H, NH); 5.13-5.01 (*m*, PhC*H*$_2$O); 4.41-4.37 (*m*, CH(2) of Pro); 3.90-3.57 (*m*, CH_2 of Gly and CH_2(5) of Pro); 2.14-1.81 (*m*, CH_2(3) and CH_2(4) of Pro); 1.48, 1.45, 1.41 (3*s*, 4 Me of 2 Aib). ^{13}C-NMR (CD_3OD): 178.5, 174.4, 173.7, 171.2 (4*s*, COOH, CO); 159.0 (*s*, CO(urethane)); 67.7 (*t*, PhC*H*$_2$O); 63.8 (*d*, C(2) of Pro); 57.6, 57.1 (2*s*, 2 C(2) of 2 Aib); 49.5, 44.2 (2*t*, C(5) of Pro and C(2) of Gly); 29.5, 26.4 (2*t*, C(3) and C(4) of Pro); 26.1, 26.0, 24.5, 24.3 (4*q*, 4 Me of 2 Aib). ESI-MS (MeOH+NaI): 499 (100, [*M*+Na]$^+$). Anal. calc. for $C_{23}H_{32}N_4O_7$•1/3H_2O (482.53): C 57.25, H 6.82, N 11.61; found: C 57.28, H 6.72, N 11.37.

7.3. *Z-Gly-Aib-Pro-Aib-Phe-Aib-OtBu* (**21**). Z-Phe-Aib-O*t*Bu (**20** [15], 0.25 g, 0.57 mmol) was hydrogenated according to the *GP 3* (H_2, 30 mg Pd/C, 10 ml MeOH). Reaction time: 24 h. Purification by CC (AcOEt/MeOH 12:1) yielded 0.168 g (97%) of the *N*-deprotected dipeptide, which was used directly in the next step.

The coupling of **19** (0.24 g, 0.5 mmol) with the above prepared dipeptide H-Phe-Aib-O*t*Bu (0.168 g, 0.55 mmol) in abs. CH_2Cl_2 (8 ml) and abs. MeCN (2 ml) was performed using PyAOP (0.262 g, 0.55 mmol) and DIEA (0.129 g, 1.0 mmol) according to the *GP 4*. Reaction time: 20 h. Purification by CC (AcOEt/MeOH 15:1), followed by prep. TLC (AcOEt/MeOH 16:1) yielded 0.213 g (56%) of **21** as a white foam. IR: 3314*s*, 3030*w*, 2983*m*, 2935*m*, 2878*w*, 1729*s*, 1662*s*, 1623*s*, 1533*s*, 1470*m*, 1455*m*, 1409*m*, 1384*m*, 1366*m*, 1271*m*, 1248*m*, 1148*s*, 1050*m*, 980*w*, 940*w*, 849*w*, 754*m*, 699*m*. ^{1}H-NMR (CD_3OD): 7.70, 7.62, 7.57 (3*s*, 3 NH); 7.34-7.12 (*m*, 10 arom. H, 2 NH); 5.08 (br.*s*, PhC*H*$_2$O); 4.55-4.45, 4.25-4.18 (2*m*, CH(2) of Pro and CH(2) of Phe); 3.95-3.52 (*m*, CH_2 of Gly

and CH_2(5) of Pro); 3.43-3.34 (*m*, 1H of CH_2 of Phe); 2.90-2.80 (*m*, 1H of CH_2 of Phe); 2.30-1.60 (*m*, CH_2(3) and CH_2(4) of Pro); 1.54, 1.47, 1.46, 1.45, 1.37, 1.12 (6*s*, 6 Me of 3 Aib and Me_3C). ^{13}C-NMR (CD_3OD): 177.3, 177.2, 175.0, 174.8, 172.8, 171.2 (6*s*, 6 CO); 159.1 (*s*, CO(urethane)); 139.3, 138.0 (2*s*, 2 arom. C); 130.0, 129.4, 129.2, 129.0, 128.7, 127.4 (6*d*, 10 arom. CH); 81.7 (*s*, Me_3C); 67.6 (*t*, PhCH_2O); 65.4 (*d*, C(2) of Pro); 58.0, 57.6, 57.5 (3*s*, 3 C(2) of 3 Aib); 56.3 (*d*, C(2) of Phe); 50.0, 44.0 (2*t*, C(5) of Pro and C(2) of Gly); 38.4 (*t*, C(3) of Phe); 29.7 (*t*, C(3) of Pro); 28.2 (*q*, Me_3C); 26.9 (*t*, C(4) of Pro); 27.2, 26.8, 25.7, 24.7, 23.84, 23.77 (6*q*, 6 Me of 3 Aib). ESI-MS (MeOH+NaI): 788 (100, $[M+Na]^+$). Anal. calc. for $C_{40}H_{56}N_6O_9 \bullet 1/2H_2O$ (773.93): C 62.08, H 7.42, N 10.86; found: C 62.19, H 7.55, N 10.85.

7.4. *Attempted Cyclization of* **21**. The N-terminus of **21** (0.193 g, 0.25 mmol) was deprotected according to the *GP 3* (H_2, 25 mg Pd/C, 6 ml MeOH): 0.145 g (0.23 mmol, 91%) H-Gly-Aib-Pro-Aib-Phe-Aib-O*t*Bu were obtained after purification by CC (CH_2Cl_2/MeOH/NH_3(l) 10:1:0.1) as a white foam. This peptide was dissolved in CH_2Cl_2/TFA (20 ml) at r.t. and stirred for 5 h. Excess TFA was removed as described in section 4.1 and, after drying i.v., 0.19 g (0.23 mmol, quant. yield) of the deprotected hexapeptide was obtained as its TFA salt.

DEPBT-mediated cyclization: According to the *GP 5*, a soln. of the linear peptide (91 mg, 0.11 mmol) in abs. DMF (80 ml) was subjected to cyclization with DEPBT (0.137 g, 0.46 mmol) and DIEA (0.8 ml). No cyclopeptide **22** was obtained after 3 d reaction time.

PyBOP-mediated cyclization: Following the *GP 7*, with 91 mg (0.11 mmol) of the linear precursor in abs. DMF (230 ml), PyBOP (0.18 g, 0.34 mmol) and DIEA (2.3 ml). No cyclic product was formed after 3 d reaction time.

8. *Synthesis of Fmoc-Pro-Aib-Gly-Aib-Phe-Aib-N(Me)Ph* (**27**).

8.1. *Fmoc-Pro-Aib-Gly-Aib-N(Me)Ph* (**24**). The N-terminus of **23** (0.385 g, 1.0 mmol) was deprotected (H_2, 40 mg Pd/C, 6 ml MeOH) according to the *GP 3*: 0.25 g (quant. yield) of H-Gly-Aib-N(Me)Ph were obtained as a pale yellow foam and used directly in the next coupling step.

Following *GP 4*, a soln. of **5** (0.385 g, 0.91 mmol) and the above-prepared dipeptide (0.25 g, 1.0 mmol) in abs. CH_2Cl_2 (6 ml) and abs. DMF (2 ml) was treated with HATU (0.38 g, 1.0 mmol), HOAt (0.136 g, 1.0 mmol, 2 ml 0.5M DMF-soln.) and DIEA (0.258 g, 2.0 mmol). Reaction time: 24 h. Purification by CC (CH_2Cl_2/MeOH 15:1, then AcOEt/MeOH 12:1) afforded 0.45 g (76%) of **24** as a white foam. IR: 3423*s*, 3063*m*, 2982*m*, 2932*m*, 1670*s*, 1593*m*, 1537*s*, 1495*s*, 1468*m*, 1451*s*, 1423*s*, 1389*m*, 1359*s*, 1337*m*, 1284*m*, 1244*m*, 1192*m*, 1171*m*, 1122*m*, 1091*m*, 1022*w*, 991*w*, 874*w*, 760*m*, 741*m*, 704*m*. ^{1}H-NMR ($CDCl_3$): 7.76, 7.74 (2br.*s*, 2 NH); 7.58-7.20 (*m*, 13 arom. H); 6.84 (br.*s*, NH); 4.42-4.08 (*m*, C*H*C*H*$_2$O of Fmoc, CH(2) of Pro); 3.86-3.48 (*m*, CH_2(5) of Pro and CH_2 of Gly); 3.26 (*s*, MeN); 2.08-1.80 (*m*, CH_2(3) and CH_2(4) of Pro); 1.50, 1.47, 1.42 (3*s*, 4 Me of 2 Aib). ^{13}C-NMR ($CDCl_3$): 173.5, 173.0, 172.5, 168.4 (4*s*, 4 CO); 156.0 (*s*, CO(urethane)); 145.1, 143.5, 141.2 (3*s*, 5 arom. C); 129.0, 127.7, 127.3, 127.1, 127.0, 124.9, 119.9 (7*d*, 13 arom. CH); 67.9 (*t*, CH*C*H$_2$O of Fmoc); 61.0 (*d*, C(2) of Pro); 57.25, 57.20 (2*s*, 2 C(2) of 2 Aib); 47.1 (*t*, C(5) of Pro); 46.9 (*d*, *C*HCH$_2$O of Fmoc); 43.3 (*t*, C(2) of Gly); 40.4 (*q*, MeN); 29.1 (*t*, C(3) of Pro); 26.6, 26.1,

24.5 (3*q*, 4 Me of 2 Aib); 24.7 (*t*, C(4) of Pro). ESI-MS (MeOH+NaI): 676 (100, [*M*+Na]$^+$). Anal. calc. for $C_{37}H_{43}N_5O_6$•1/2H_2O (662.78): C 67.05, H 6.69, N 10.56; found: C 67.10, H 6.63, N 10.50.

8.2. *Fmoc-Pro-Aib-Gly-Aib-OH* (**25**). According to *GP 2*, a soln. of **24** (0.387 g, 0.59 mmol) in 6 ml of 3N HCl (MeCN/H_2O 1:1) was stirred for 20 h. Purification by CC (CH_2Cl_2/MeOH 10:1) yielded 0.247 g (74%) of **25** as a white foam. IR: 3319*s*, 3066*m*, 2984*m*, 2940*m*, 2882*m*, 1673*s*, 1539*s*, 1470*m*, 1451*s*, 1427*s*, 1390*m*, 1357*m*, 1337*m*, 1286*m*, 1244*m*, 1213*m*, 1167*m*, 1127*m*, 1091*m*, 1043*w*, 1020*w*, 989*w*, 939*w*, 915*w*, 876*w*, 759*m*, 740*m*. ^{1}H-NMR: 8.62 (*s*, NH); 7.90-7.88 (*m*, 2 arom. H); 7.78 (*t*, *J* = 5.9, NH of Gly); 7.71-7.31 (*m*, 6 arom. H, NH); 4.43-4.14 (*m*, C*H*C*H*$_2$O of Fmoc, CH(2) of Pro); 3.67-3.32 (*m*, CH_2 of Gly, CH_2(5) of Pro); 2.30-1.80 (*m*, CH_2(3) and CH_2(4) of Pro); 1.37, 1.33, 1.31, 1.28 (4*s*, 4 Me of 2 Aib). ^{13}C-NMR: 175.1, 173.7, 172.7, 168.1 (4*s*, 4 CO); 154.2 (*s*, CO(urethane)); 143.7, 143.5, 140.6 (3*s*, 4 arom. C); 127.5, 127.0, 125.3, 125.2, 125.0, 120.0 (6*d*, 8 arom. CH); 66.6 (*t*, CH*C*H$_2$O of Fmoc); 59.6 (*d*, C(2) of Pro); 55.9, 54.5 (2*s*, 2 C(2) of 2 Aib); 47.1 (*t*, C(5) of Pro); 46.5 (*d*, *C*HCH$_2$O of Fmoc); 42.6 (*t*, C(2) of Gly); 29.3 (*t*, C(3) of Pro); 25.6, 24.7, 24.4, 24.3 (4*q*, 4 Me of 2 Aib); 24.0 (*t*, C(4) of Pro). ESI-MS (MeOH+NaI): 587 (100, [*M*+Na]$^+$), 565 (15, [*M*+H]$^+$). Anal. calc. for $C_{30}H_{36}N_4O_7$•1/2H_2O (573.65): C 62.81, H 6.50, N 9.77; found: C 62.70, H 6.57, N 9.77.

8.3. *Fmoc-Pro-Aib-Gly-Aib-Phe-Aib-N(Me)Ph* (**27**). Prior to coupling, Z-Phe-Aib-N(Me)Ph (**26**) [16] (0.515 g, 1.09 mmol) was hydrogenated according to the *GP 3* (H_2, 60 mg Pd/C, 10 ml MeOH). Purification by CC (AcOEt/MeOH

15:1) yielded 0.365 g (99%) H-Phe-Aib-N(Me)Ph as a white foam, which was used directly in the next step.

The coupling of **25** (0.198 g, 0.35 mmol) with the above-prepared dipeptide (0.13 g, 0.38 mmol) in abs. CH_2Cl_2 (8 ml) and abs. MeCN (2 ml) was performed using TBTU (0.128 g, 0.4 mmol), HOBt (0.054 g, 0.4 mmol) and DIEA (0.103 g, 0.8 mmol) according to the *GP 4*. Reaction time: 48 h. Purification by CC (CH_2Cl_2/MeOH 20:1, then AcOEt/MeOH/hexane 8:1:0.5) yielded 0.237 g (76%) of **27** as a white foam. IR: 3423*s*, 3060*m*, 3025*m*, 2980*m*, 2936*m*, 1665*s*, 1594*m*, 1533*s*, 1496*s*, 1470*m*, 1453*s*, 1424*m*, 1386*m*, 1361*m*, 1338*m*, 1277*m*, 1244*m*, 1191*m*, 1121*m*, 1091*m*, 758*w*, 741*m*, 704*m*. ^{1}H-NMR (CD_3OD): 7.81-7.78, 7.62-7.59, 7.41-7.06 (3*m*, 18 arom. H, 5 NH); 4.42-4.17 (*m*, C*H*C*H*$_2$O of Fmoc, CH(2) of Phe and CH(2) of Pro); 3.74-3.45 (*m*, CH_2 of Gly, CH_2(5) of Pro and 1H of CH_2 of Phe); 3.26 (*s*, MeN); 2.98-2.80 (*m*, 1H of CH_2 of Phe); 2.20-1.80 (*m*, CH_2(3) and CH_2(4) of Pro); 1.50, 1.49, 1.45, 1.43, 1.27, 1.11 (6*s*, 6 Me of 3 Aib). ^{13}C-NMR (CD_3OD): 178.0, 176.3, 175.3, 174.9, 172.5, 171.9 (6*s*, 6 CO); 157.0 (*s*, CO(urethane)); 146.6, 145.3, 145.1, 142.5, 139.2 (5*s*, 6 arom. C); 130.3, 129.2, 128.8, 128.1, 127.4, 126.1, 126.0, 121.0 (8*d*, 18 arom. CH); 68.8 (*t*, CH*C*H$_2$O of Fmoc); 61.9 (*d*, C(2) of Pro); 58.3, 57.9, 57.7 (3*s*, 3 C(2) of 3 Aib); 61.9, 56.4 (2*d*, C(2) of Pro and C(2) of Phe); 48.3 (*d*, *C*HCH$_2$O of Fmoc); 48.1, 45.2 (2*t*, C(5) of Pro and C(2) of Gly); 41.2 (*q*, MeN); 37.7 (*t*, C(3) of Phe); 31.0 (*t*, C(3) of Pro); 26.6, 25.6, 25.2 (3*q*, 6 Me of 3 Aib); 25.5 (*t*, C(4) of Pro). ESI-MS (MeOH+NaI): 909 (100, $[M+Na]^+$). Anal. calc. for $C_{50}H_{59}N_7O_8 \bullet 1/3H_2O$ (892.06): C 67.32, H 6.74, N 10.99; found: C 67.32, H 6.78, N 11.04.

8.4. *Attempted Cyclization of* **27**. According to the *GP 2*, a soln. of **27** (0.222 g, 0.25 mmol) in 4 ml of 3N HCl (MeCN/H_2O 1:1) was stirred for 24 h. The product was extracted from the mixture with CH_2Cl_2, dried (Na_2SO_4), and concentrated: 0.195 g (98%) peptide acid was obtained as a white foam after drying i.v. This acid was then dissolved in CH_2Cl_2 (5 ml) and Et_2NH (1 ml) was added. After 4 h of stirring at r.t., the solvents were removed under reduced pressure. The product was triturated with Et_2O, collected by filtration and washed with Et_2O to give 0.13 g (93%) of deprotected hexapeptide as a pale yellow foam, which was used in the cyclization without further purification.

This peptide (0.13 g, 0.23 mmol) was dissolved in 450 ml of abs. CH_2Cl_2/DMF (2:1) and subjected to cyclization with PyAOP (0.59 g, 1.13 mmol), HOAt (1.13 mmol, 2.5 ml 0.5M DMF-soln.) and DIEA (4.5 ml) by following the *GP 7*. No cyclic product was obtained after 3 d reaction time. By purification on *Sephadex LH-20*, eluting with MeOH, 90 mg (0.16 mmol) of starting material was recovered. The recovered linear peptide (90 mg, 0.16 mmol) was then dissolved in abs. DMF (160 ml) and subjected to cyclization according to the *GP 5*, using DEPBT (0.233 g, 0.78 mmol) and DIEA (1.6 ml). No cyclic product was formed after 3 d reaction time.

9. *Synthesis of Z-Gly-Aib-Aib-Pro-Aib-Aib-OtBu* (**30**).

9.1. *Fmoc-Pro-Aib-Aib-OtBu* (**29**). According to the *GP 4*, the coupling of **5** (0.414 g, 0.98 mmol) with HCl•H-Aib-O*t*Bu (0.211 g, 1.08 mmol) was carried out in abs. CH_2Cl_2 (8 ml) and abs. MeCN (2 ml) using PyBOP (0.51 g, 0.98 mmol) and DIEA (0.388 g, 3.0 mmol). Reaction time: 24 h. Purification by CC (CH_2Cl_2/MeOH 35:1, then AcOEt/hexane 2:1) afforded 0.459 g (83%) of pure **29**.

White powder. M.p. 153-154°. IR: 3416*m*, 3281*m*, 3041*w*, 2981*m*, 2942*m*, 2876*m*, 1731*s*, 1710*s*, 1687*s*, 1648*s*, 1530*s*, 1470*m*, 1450*s*, 1416*s*, 1389*m*, 1360*s*, 1331*m*, 1307*m*, 1242*m*, 1152*s*, 1097*m*, 1033*w*, 991*w*, 939*w*, 855*w*, 760*m*, 739*m*. ^{1}H-NMR: 8.21 (*s*, NH); 7.91-7.89 (*m*, 2 arom. H); 7.64-7.61 (*m*, 2 arom. H); 7.43-7.32 (*m*, 4 arom. H); 7.20 (*s*, NH); 4.32-4.13 (*m*, C*H*C*H*$_2$O of Fmoc and CH(2) of Pro); 3.47-3.38 (*m*, CH$_2$(5) of Pro); 2.20-1.80 (*m*, CH$_2$(3) and CH$_2$(4) of Pro); 1.37, 1.32, 1.31, 1.22 (4*s*, 4 Me of 2 Aib and Me$_3$C). ^{13}C-NMR: 172.7, 171.2 (2*s*, 3 CO); 154.1 (*s*, CO(urethane)); 143.6, 143.5, 140.6 (3*s*, 4 arom. C); 127.6, 127.0, 124.9, 120.0 (4*d*, 8 arom. CH); 79.0 (*s*, Me$_3$*C*); 66.5 (*t*, CH*C*H$_2$O of Fmoc); 59.8 (*d*, C(2) of Pro); 55.7, 55.2 (2*s*, 2 C(2) of 2 Aib); 46.5 (*d*, *C*HCH$_2$O of Fmoc); 46.4 (*t*, C(5) of Pro); 29.4 (*t*, C(3) of Pro); 27.3 (*q*, *Me*$_3$C); 26.2, 24.8, 23.7, 23.6 (4*q*, 4 Me of 2 Aib); 24.0 (*t*, C(4) of Pro). ESI-MS (MeOH+NaI): 586 (100, [*M*+Na]$^+$). Anal. calc. for $C_{32}H_{41}N_3O_6$ (563.69): C 68.18, H 7.33, N 7.45; found: C 67.85, H 7.34, N 7.41.

9.2. *Z-Gly-Aib-Aib-Pro-Aib-Aib-O*t*Bu* (**30**). Prior to coupling, **29** (0.382 g, 0.68 mmol) was deprotected with Et$_2$NH (1 ml) in CH$_2$Cl$_2$ (5 ml) by stirring for 6 h. The solvents were then removed under reduced pressure and the residue purified by CC (CH$_2$Cl$_2$/MeOH/NH$_3$(l) 11:1:0.1) to afford 0.172 g (74%) of H-Pro-Aib-Aib-O*t*Bu, which was used directly in the next step.

A soln. of Z-Gly-Aib-Aib-OH (0.19 g, 0.5 mmol) and the above-prepared tripeptide (0.172 g, 0.5 mmol) in abs. MeCN (8 ml) was treated with PyBOP (0.29 g, 0.56 mmol) and DIEA (0.143 g, 1.1 mmol) according to the *GP 4*. Reaction time: 24 h. Purification by CC (AcOEt/MeOH 10:1, then CH$_2$Cl$_2$/MeOH 15:1) yielded 0.248 g (64%) pure **30** as a white foam. IR: 3336*s*, 3033*m*, 2983*s*, 2937*m*,

2877*w*, 1658*s*, 1627*s*, 1521*s*, 1469*s*, 1455*s*, 1413*m*, 1384*m*, 1366*m*, 1305*m*, 1276*s*, 1234*s*, 1148*s*, 1048*m*, 987*w*, 939*w*, 850*w*, 751*w*, 698*m*. ^{1}H-NMR (CD_3OD): 7.54 (br.*s*, NH); 7.36-7.28 (*m*, 5 arom. H, 4 NH); 5.13 (br.*s*, PhCH_2O); 4.20 (*t*, *J* = 7.9, CH(2) of Pro); 3.71-3.62 (*m*, CH_2 of Gly and CH_2(5) of Pro); 2.30-1.65 (*m*, CH_2(3) and CH_2(4) of Pro); 1.51, 1.49, 1.45, 1.42, 1.40 (5*s*, 8 Me of 4 Aib and Me_3C). ^{13}C-NMR (CD_3OD): 176.3, 176.0, 175.3, 174.9, 174.4, 171.6 (6*s*, 6 CO); 159.0 (*s*, CO(urethane)); 138.2 (*s*, 1 arom. C); 129.5, 128.9, 128.4 (3*d*, 5 arom. CH); 81.5 (*s*, Me_3C); 67.6 (*t*, PhCH_2O); 64.8 (*d*, C(2) of Pro); 57.9, 57.8, 57.45, 57.36 (4*s*, 4 C(2) of 4 Aib); 49.7, 45.5 (2*t*, C(5) of Pro and C(2) of Gly); 29.7 (*t*, C(3) of Pro); 28.1 (*q*, Me_3C); 26.9 (*t*, C(4) of Pro); 27.4, 25.9, 25.3, 24.2, 24.1 (6*s*, 8 Me of 4 Aib). ESI-MS (MeOH+NaI): 741 (15, $[M+K]^+$), 725 (100, $[M+Na]^+$), 703 (100, $[M+H]^+$). Anal. calc. for $C_{35}H_{54}N_6O_9 \bullet 1/2\ H_2O$ (708.85): C 59.30, H 7.77, N 11.85; found: C 59.07, H 7.78, N 11.83.

9.3. *Attempted Cyclization of* **30**. According to the *GP 3* (H_2, 20 mg Pd/C, 6 ml MeOH), the N-terminus of **30** (0.188 g, 0.27 mmol) was deprotected to give, after purification by CC (CH_2Cl_2/MeOH/NH_3(l) 11:1:0.1), 0.146 g (97%) H-Gly-Aib-Aib-Pro-Aib-Aib-O*t*Bu as a white foam. This foam was then dissolved in 22 ml of CH_2Cl_2/TFA (1:1) and stirred for 5 h at r.t. Excess TFA was removed as described in section 4.1 and 0.17 g (0.26 mmol) of deprotected hexapeptide was obtained as its TFA salt after drying i.v.. This material (0.26 mmol) was dissolved in 170 ml abs. DMF and subjected to cyclization with DEPBT (0.308 g, 1.03 mmol) and DIEA (1.7 ml) by following the *GP 5*. No reaction was observed after 3 d, and the linear precursor (not as its TFA salt) was fully recovered from the mixture by purification on *Sephadex LH-20*, eluting with MeOH. The peptide

(0.134 g, 0.26 mmol) was then subjected to cyclization with DEPC (0.17 g, 1.04 mmol) and DIEA (1.9 ml) in 190 ml abs. DMF according to the *GP 6*. However, no cyclic product was formed after 10 d reaction time.

10. *Synthesis of Z-Phe-Pro-Aib-Pro-Aib-Aib-N(Me)Ph* (**33**).

10.1. *Fmoc-Pro-Aib-Aib-N(Me)Ph* (**32**). According to the *GP 1*, **5** (0.484 g, 1.14 mmol) in THF (15 ml) was treated with **3** (0.22 g, 1.26 mmol) in THF (1 ml). Reaction time: 6 d. Purification by CC (AcOEt → AcOEt/MeOH 10:1) gave 0.475 g (70%) of **32**. White powder. M.p. 151.0-151.8°. IR: 3384*s*, 3303*s*, 3062*w*, 3039*w*, 2983*m*, 2945*m*, 2890*w*, 1712*s*, 1689*s*, 1648*s*, 1594*m*, 1539*s*, 1494*s*, 1450*s*, 1423*s*, 1392*m*, 1360*s*, 1334*m*, 1285*m*, 1242*m*, 1215*m*, 1203*m*, 1165*m*, 1122*m*, 1088*m*, 1033*w*, 1022*w*, 991*w*, 877*w*, 759*m*, 740*m*, 707*m*. ^{1}H-NMR ($CDCl_3$): 7.77-7.19 (*m*, 13 arom. H, NH); 6.83 (br.*s*, NH); 4.42-4.17 (*m*, $CHCH_2O$ of Fmoc, CH(2) of Pro); 3.64-3.47 (*m*, CH_2(5) of Pro); 3.24 (*s*, MeN); 2.20-1.89 (*m*, CH_2(3) and CH_2(4) of Pro); 1.48, 1.45, 1.42, 1.41 (4*s*, 4 Me of 2 Aib). ^{13}C-NMR ($CDCl_3$): 173.5, 172.6, 171.1 (3*s*, 3 CO); 156.4 (*s*, CO(urethane)); 144.5, 143.7, 141.2 (3*s*, 5 arom. C); 129.2, 128.0, 127.7, 127.0, 124.9, 119.9 (6*d*, 13 arom. CH); 67.6 (*t*, $CHCH_2O$ of Fmoc); 61.2 (*d*, C(2) of Pro); 58.1, 57.4 (2*s*, 2 C(2) of 2 Aib); 47.1 (*d*, $CHCH_2O$ of Fmoc); 46.9 (*t*, C(5) of Pro); 41.0 (*q*, MeN); 28.7 (*t*, C(3) of Pro), 25.4, 24.7 (2*q*, 4 Me of 2 Aib); C(4) of Pro not detectable. ESI-MS (MeOH+NaI): 619 (100, $[M+Na]^+$). Anal. calc. for $C_{35}H_{40}N_4O_5$ (596.73): C 70.45, H 6.76, N 9.39; found: C 70.35, H 6.75, N 9.32.

10.2. *Z-Phe-Pro-Aib-Pro-Aib-Aib-N(Me)Ph* (**33**). The Fmoc group of **32** (0.375 g, 0.63 mmol) was removed with Et_2NH (2 ml) in CH_2Cl_2 (8 ml) as described in section 9.2. Reaction time: 5 h. Purification by CC

(CH_2Cl_2/MeOH/NH_3(l) 12:1:0.1) yielded 0.22 g (94%) of *N*-deprotected tripeptide as a white foam which was used directly in the next step.

The coupling of **11** (0.257 g, 0.53 mmol) with the above-prepared H-Pro-Aib-Aib-N(Me)Ph (0.22 g, 0.59 mmol) was accomplished using HATU (0.223 g, 0.59 mmol), HOAt (80 mg, 0.59 mmol, 1.2 ml of 0.5M DMF-soln.) and DIEA (0.142 g, 1.07 mmol) in MeCN (6 ml) according to the *GP 4*. Reaction time: 24 h. Purification by CC (AcOEt/MeOH 12:1) yielded 0.312 g (70%) **33** as a white foam. IR: 3422*s*, 3321*s*, 3061*w*, 3030*m*, 2983*m*, 2940*m*, 2876*m*, 1718*s*, 1659*s*, 1594*m*, 1532*s*, 1496*s*, 1453*s*, 1416*s*, 1391*m*, 1362*m*, 1282*m*, 1244*m*, 1213*m*, 1173*m*, 1089*m*, 1053*m*, 1028*m*, 925*w*, 880*w*, 767*w*, 749*w*, 702*m*. ^{1}H-NMR (CD_3OD): 7.78, 7.68 (2br.*s*, 2 NH); 7.36.7.19 (*m*, 15 arom. H, 2 NH); 4.99 (br.*s*, PhC*H*$_2$O); 4.64-4.13 (*m*, CH(2) of Phe, 2 CH(2) of 2 Pro); 3.94-3.38 (*m*, 2 CH_2(5) of 2 Pro); 3.33 (br.*s*, MeN); 3.04-2.71 (*m*, CH_2 of Phe); 2.27-1.67 (*m*, 2 CH_2(3) and 2 CH_2(4) of 2 Pro); 1.54, 1.52, 1.50, 1.49, 1.45 (5*s*, 6 Me of 3 Aib). ^{13}C-NMR (CD_3OD): 176.6, 175.6, 174.7, 174.5, 173.7, 172.6 (6*s*, 6 CO); 158.2 (*s*, CO(urethane)); 146.9, 138.1, 137.6 (3*s*, 3 arom. C); 130.2, 130.1, 129.4, 129.3, 128.9, 128.6, 128.1, 127.9, 127.7 (9*d*, 15 arom. CH); 67.5 (*t*, Ph*C*H$_2$O); 64.8, 61.4 (2*d*, 2 C(2) of 2 Pro); 58.4, 58.3, 57.6 (3*s*, 3 C(2) of 3 Aib); 55.7 (*d*, C(2) of Phe); 49.8, 48.6 (2*t*, 2 C(5) of 2 Pro); 41.0 (*q*, MeN); 38.3 (*t*, C(3) of Phe); 30.5, 29.6 (2*t*, 2 C(3) of 2 Pro); 27.2 (*q*, Me of Aib); 27.0 (*t*, C(4) of Pro); 26.3, 26.2 (2*q*, 2 Me of Aib); 26.1 (*t*, C(4) of Pro); 26.0, 24.9, 24.4 (3*q*, 3 Me of Aib). ESI-MS (MeOH+NaI): 861 (100, $[M+Na]^+$).

10.3. *Attempted Cyclization of* **33**. Hexapeptide **33** (0.255 g, 0.3 mmol) was hydrolyzed according to the *GP 2* with 4 ml of 3N HCl (MeCN/H_2O 1:1).

Reaction time: 5 h. The product was extracted with CH_2Cl_2, dried (Na_2SO_4), concentrated and dried u.h.v. to give 0.212 g (93%) of peptide acid as a white foam, which was used in the next step without further purification. The Z protecting group was then removed according to the *GP 3* (H_2, 23 mg Pd/C, 10 ml MeOH). Reaction time: 16 h; 0.172 g of the deprotected linear precursor were obtained as a pale yellow foam.

A soln. of 86 mg (0.14 mmol) of the above-prepared linear precursor in 140 ml of abs. DMF was subjected to cyclization with PyAOP (0.292 g, 0.56 mmol), HOAt (0.56 mmol, 1.2 ml 0.5M DMF-soln.) and DIEA (1.4 ml) according to the *GP 7*. No cyclic product **34** was obtained after 3 d reaction time.

The linear precursor (86 mg, 0.14 mmol) was dissolved in abs. DMF (140 ml) under stirring. The mixture was cooled to 0° and EDCI•HCl (0.134 g, 0.7 mmol), HOAt (0.7 mmol, 1.4 ml 0.5M DMF-soln.) and DIEA (1.4 ml) were added. After 2 h at 0°, the mixture was warmed to r.t. and stirred for an additional 3 d. No cyclic product **34** could be isolated.

11. *X-Ray Crystal-Structure Determination of* **1**•Et(*i*-Pr)$_2$NH$^+$PF$_6^-$•H_2O (see *Table 3* and *Figure 1*)[4]). All measurements were made on a *Nonius KappaCCD* area-detector diffractometer [24] using graphite-monochromated MoK_α radiation (λ0.71073 Å) and an *Oxford Cryosystems Cryostream 700* cooler. Data reduction was performed with *HKL Denzo and Scalepack* [25]. The

[4]) CCDC-246353 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge via www.ccdc.cam.ac.uk/conts/retrieving.html (or from the Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge CB2 1EZ, UK; fax: +44 1223 336033; e-mail: deposit@ccdc.cam.ac.uk).

intensities were corrected for *Lorentz* and polarization effects, but not for absorption. Data collection and refinement parameters are given in *Table 3*, and a view of the molecule is shown in *Figure 1*. The structure was solved by direct methods using SIR92 [26], which revealed the positions of all non-H-atoms. The asymmetric unit contains one molecule of the peptide **1**, one H_2O molecule and one unit of ethyl(diisopropyl)ammonium hexafluorophosphate. The PF_6- anion is highly disordered. Three sets of P- and F-atoms were defined for this anion and refinement of the site occupation factors of each set led to values of 0.648(3), 0.157(4) and 0.195(4). The P-F and F···F distances were restrained tightly, so as to maintain octahedral geometry and uniform P-F bond lengths. Finally, neighboring atoms within and between each conformation of the disordered cation and anion were restrained to have similar atomic displacement parameters. The non-H-atoms were refined anisotropically. The H-atoms of the H_2O molecule were placed in the positions indicated by a difference electron density map and their positions were allowed to refine together with individual isotropic displacement parameters, while restraining the O-H and H···H distances to reasonable values. All remaining H-atoms were placed in geometrically calculated positions and refined using a riding model where each H-atom was assigned a fixed isotropic displacement parameter with a value equal to $1.2U_{eq}$ of its parent atom ($1.5U_{eq}$ for the Me groups). Refinement of the structure was carried out on F^2 using full-matrix least-squares procedures, which minimised the function $\Sigma w(F_o^2-F_c^2)^2$. A correction for secondary extinction was applied. Nine reflections, whose intensities were considered to be extreme outliers, were omitted from the final refinement. The absolute configuration of the peptide molecule could not be

confirmed crystallographically, as refinement of the absolute structure factor yielded an inconclusively imprecise value of 0.08(15). Therefore, all equivalent reflections, including the Friedel pairs, were merged and the peptide enantiomer used in the refinement was chosen to give the S-configuration at each chiral centre in the peptide molecule which was known from the synthesis of the compound. Neutral atom scattering factors for non-H-atoms were taken from [27], and those for H-atoms were taken from [28]. Anomalous dispersion effects were included in F_c [29]; the values for f' and f'' were those of [30]. The values of the mass attenuation coefficients are those of [31]. All calculations were performed using the SHELXL97 program [32].

In **1**, each NH group of the peptide molecule acts as a donor for H-bonds. Two of the interactions, N(1)-H and N(10)-H, are intramolecular H-bonds with the amide O-atoms that are diagonally opposed in the peptide ring. Each of these interactions has the graph set motif [33] of S(10). This corresponds with the usual spacing along the peptide chain between the donor and acceptor atoms in open chain peptide turns. N(7)-H forms a H-bond with an F-atom in each of the disordered orientations of the PF_6^- anion; graph set motif D. N(13)-H forms an intermolecular H-bond with the amide O-atom of the peptide unit adjacent to the five-membered ring of a neighboring peptide molecule and thereby links the peptide molecules into extended chains which run parallel to the *x*-axis and have a graph set motif of C(10). N(16)-H forms an intermolecular H-bond with the O-atom of the H_2O molecule. In turn, the H_2O molecule donates H-bonds to amide O-atoms in two different neighboring peptide molecules. One of these interactions embeds the H_2O molecule between two peptide molecules in the same chain

created by the N(13)-H···O H-bond. The other interaction also involves these chains, but with the sense of the interaction running along the chain in the opposite direction. The chains incorporating H_2O and peptide molecules alternately, have binary graph set motifs of $C_2^2(13)$ and $C_2^2(12)$. The cation forms an intermolecular H-bond via N(37)-H with an amide O-atom of an adjacent peptide molecule; graph set motif D. Finally, most amide NH groups in the peptide molecule have a close intramolecular 'sideways' contact with an adjacent amide N-atom. These interactions have very sharp N-H···O angles and therefore may be simply a result of the geometrical arrangement within the molecule, rather than being true H-bonds.

Table 3

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Legends

Fig.1 *ORTEP Plot* [21] *of the molecular structure of* **1**. *The co-crystallized* Et(*i*-Pr)$_2$NH$^+$PF$_6^-$ *and* H_2O *are not shown* (50% probability ellipsoids, arbitrary numbering of the atoms)

Table 1. *Torsion Angles* [°] *for* Cyclo*(Pro-Aib-Gly-Aib-Aib-Phe)* (**1**)

Residue	ϕ	ψ	ω
Pro[1]	-54.7(4)	-35.4(5)	+179.5(3)
Aib[2]	-65.1(5)	-13.8(5)	-165.9(4)
Gly[3]	+118.6(4)	+169.5(3)	+173.0(4)
Aib[4]	+53.9(5)	+46.7(5)	+173.1(3)
Aib[5]	+80.2(5)	+10.1(5)	+170.6(4)
Phe[6]	-122.0(4)	+168.0(3)	+176.9(3)

Table 2. *Hydrogen Bonding Geometry for Cyclo(Pro-Aib-Gly-Aib-Aib-Phe)* **1**.

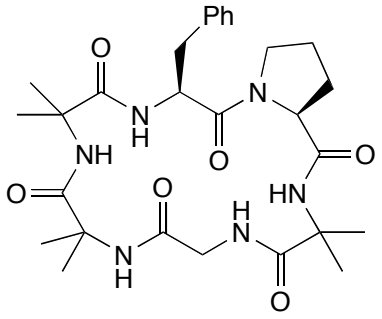
Donor (D-H)	Acceptor (A)	D-H [Å]	H···A [Å]	D···A [Å]	D-H···A [°]
N(1)-H(1)	O(12)	0.88	2.25	3.063 (5)	153
N(1)-H(1)	N(16)	0.88	2.23	2.684 (5)	112
N(7)-H(7)	N(4)	0.88	2.43	2.793 (5)	105
N(7)-H(7)	F(3a)	0.88	2.42	3.220 (5)	152
N(7)-H(7)	F(2b)	0.88	2.43	2.904 (9)	114
N(7)-H(7)	F(3c)	0.88	2.20	2.916 (8)	139
N(10)-H(10)	O(3)	0.88	2.01	2.852 (4)	159
N(10)-H(10)	N(7)	0.88	2.32	2.748 (5)	110
N(13)-H(13)	O(6^{i})	0.88	2.08	2.924 (5)	159
N(16)-H(16)	N(13)	0.88	2.53	2.868 (5)	104
N(16)-H(16)	O(44)	0.88	2.18	3.032 (5)	162
N(37)-H(37)	O(18^{ii})	0.93	1.92	2.829 (5)	165
O(44)-H(441)	O(6^{i})	0.84 (1)	1.97 (1)	2.808 (6)	173 (10)
O(44)-H(442)	O(9^{iii})	0.84 (1)	2.05 (4)	2.842 (5)	156 (9)

Superscripts in atom labels refer to the peptide molecule in the following symmetry-related positions:

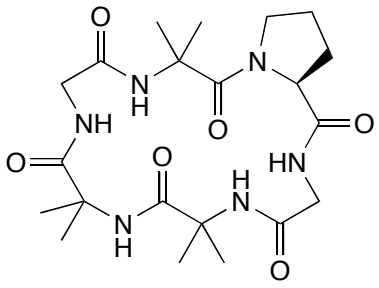
^{i} –1/2+x, -1/2+y, -z; ^{ii} –1/2+x, 1/2-y, -z; ^{iii} 1/2+x, -1/2-y, -z.

Table 3. *Crystallographic Data of* **1**•Et(*i*-Pr)$_2$NH$^+$PF$_6$-•H$_2$O.

Crystallized from		AcOEt / EtOH / hexane
Empirical formula		$C_{36}H_{62}F_6N_7PO_7$
Formula weight [g mol^{-1}]		849.89
Crystal color, habit		colorless, plate
Crystal dimensions [mm]		0.05 × 0.15 × 0.30
Temperature [K]		160(1)
Crystal system		orthorhombic
Space group		$P2_12_12_1$
Z		4
Reflections for cell determination		4318
2θ range for cell determination [°]		4-50
Unit cell parameters	a [Å]	8.7953(1)
	b [Å]	18.4796(2)
	c [Å]	26.6501(4)
	V [Å^3]	4331.54(9)
D_x [g cm^{-3}]		1.303
μ(MoK_α) [mm^{-1}]		0.142
Scan type		ϕ and ω
2θ(max) [°]		50
Total reflections measured		45195
Symmetry independent reflections		4296
Reflections with $I>2\sigma(I)$		3478
Reflections used in refinement		4287
Parameters refined; restraints		668; 997
Final $R(F)$ [$I>2\sigma(I)$ reflections]		0.0549
$wR(F^2)$ (all data)		0.1547
Weights: $w = [\sigma^2(F_o^2) + (0.0900P)^2 + 2.2715P]^{-1}$; where $P = (F_o^2+2F_c^2)/3$		
Goodness of fit		1.049
Secondary extinction coefficient		0.004(1)
Final Δ_{max}/σ		0.012
$\Delta\rho$ (max; min) [e Å^{-3}]		0.51; -0.27

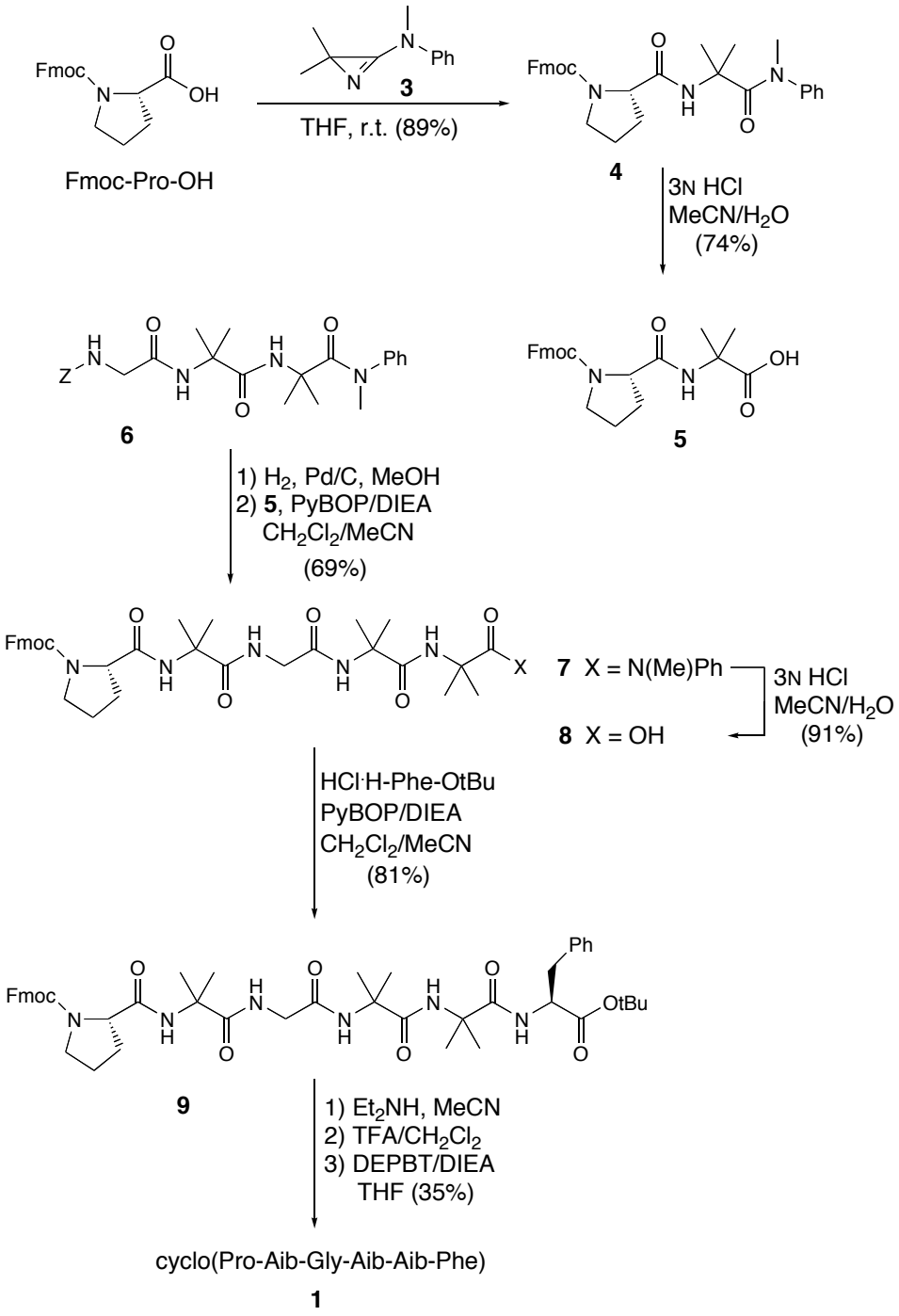


1

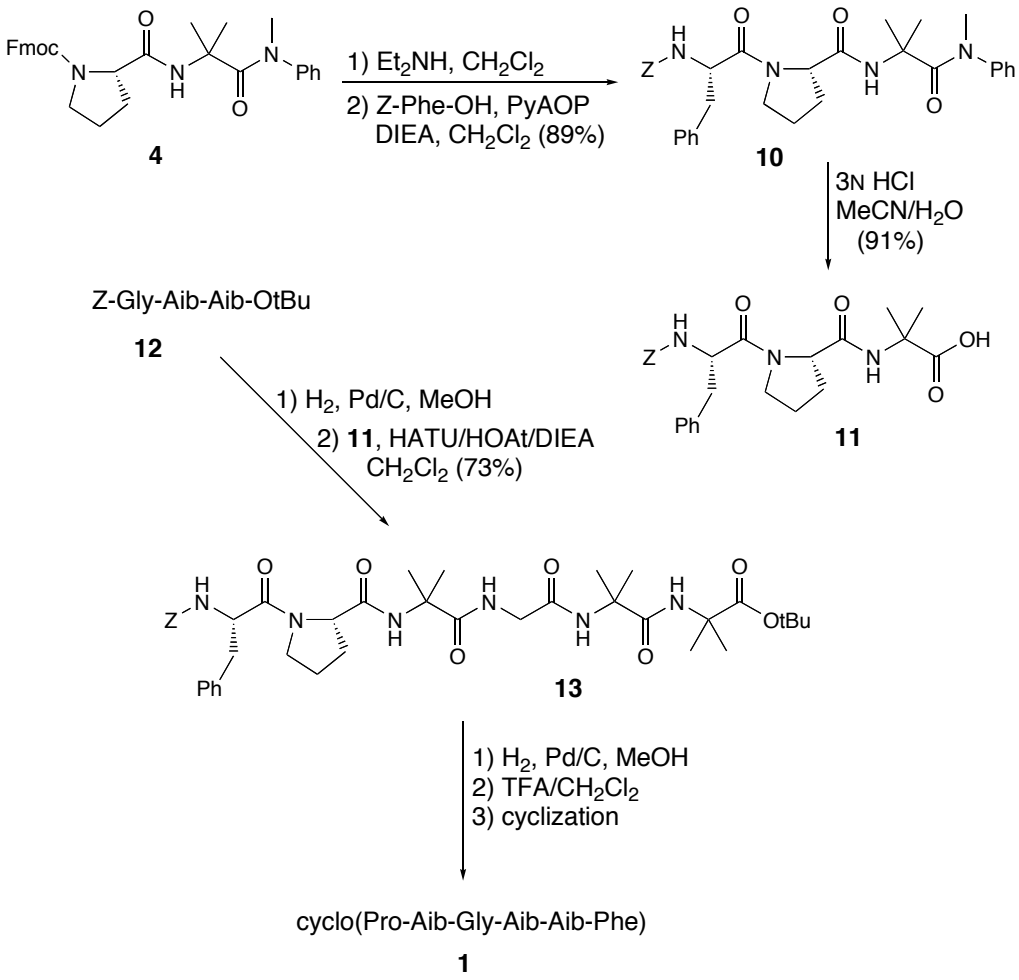


2

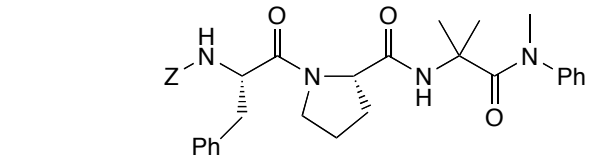
Scheme 1


Fmoc-Pro-OH
3
THF, r.t. (89%)
4
3N HCl
MeCN/H2O
(74%)
5
6
1) H2, Pd/C, MeOH
2) 5, PyBOP/DIEA
CH2Cl2/MeCN
(69%)
7 X = N(Me)Ph
8 X = OH
3N HCl
MeCN/H2O
(91%)
HCl·H-Phe-OtBu
PyBOP/DIEA
CH2Cl2/MeCN
(81%)
9
1) Et2NH, MeCN
2) TFA/CH2Cl2
3) DEPBT/DIEA
THF (35%)
cyclo(Pro-Aib-Gly-Aib-Aib-Phe)
1

Scheme 2

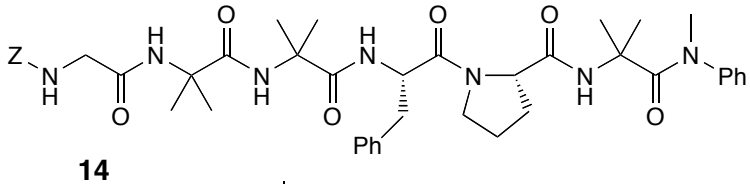

Fmoc
Ph
4
1) Et2NH, CH2Cl2
2) Z-Phe-OH, PyAOP
DIEA, CH2Cl2 (89%)
Z
Ph
10
3N HCl
MeCN/H2O
(91%)
OH
11
Z-Gly-Aib-Aib-OtBu
12
1) H2, Pd/C, MeOH
2) 11, HATU/HOAt/DIEA
CH2Cl2 (73%)
OtBu
13
1) H2, Pd/C, MeOH
2) TFA/CH2Cl2
3) cyclization
cyclo(Pro-Aib-Gly-Aib-Aib-Phe)
1

Scheme 3



10

1) H_2, Pd/C, MeOH
2) Z-Gly-Aib-Aib-OH
PyAOP/HOAt/DIEA
CH_2Cl_2 (90%)



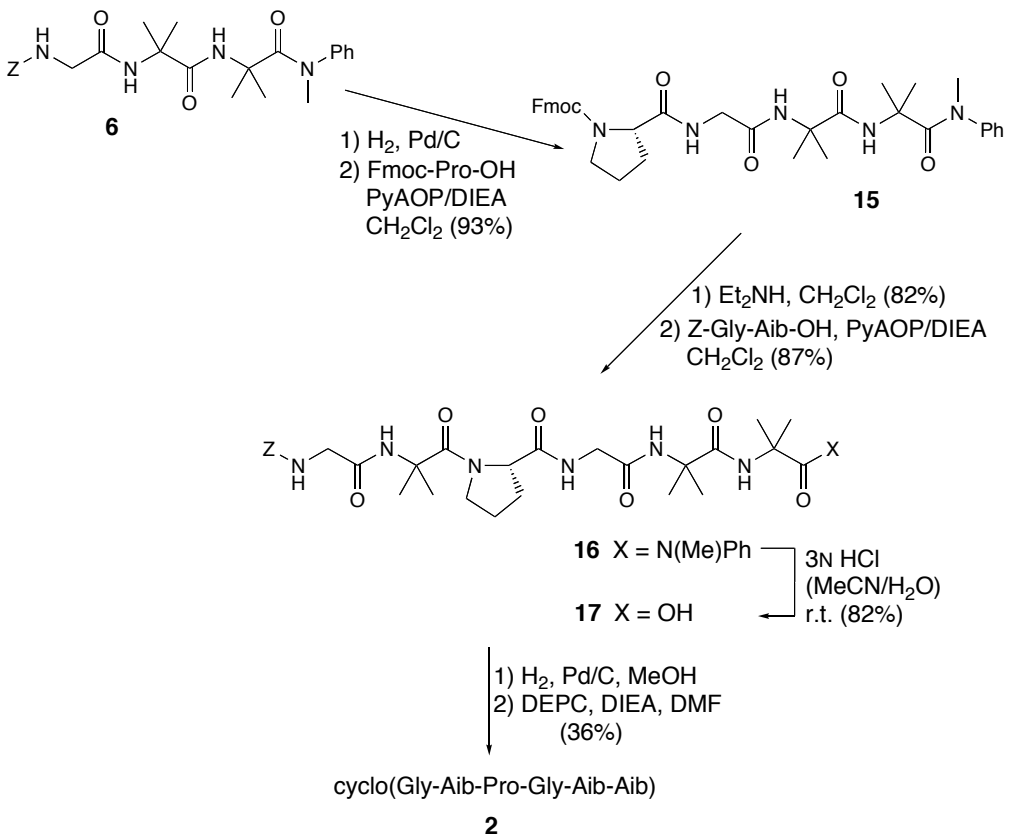
14

1) 3N HCl (MeCN/H_2O), (89%)
2) H_2, Pd/C, MeOH (95%)
3) cyclization

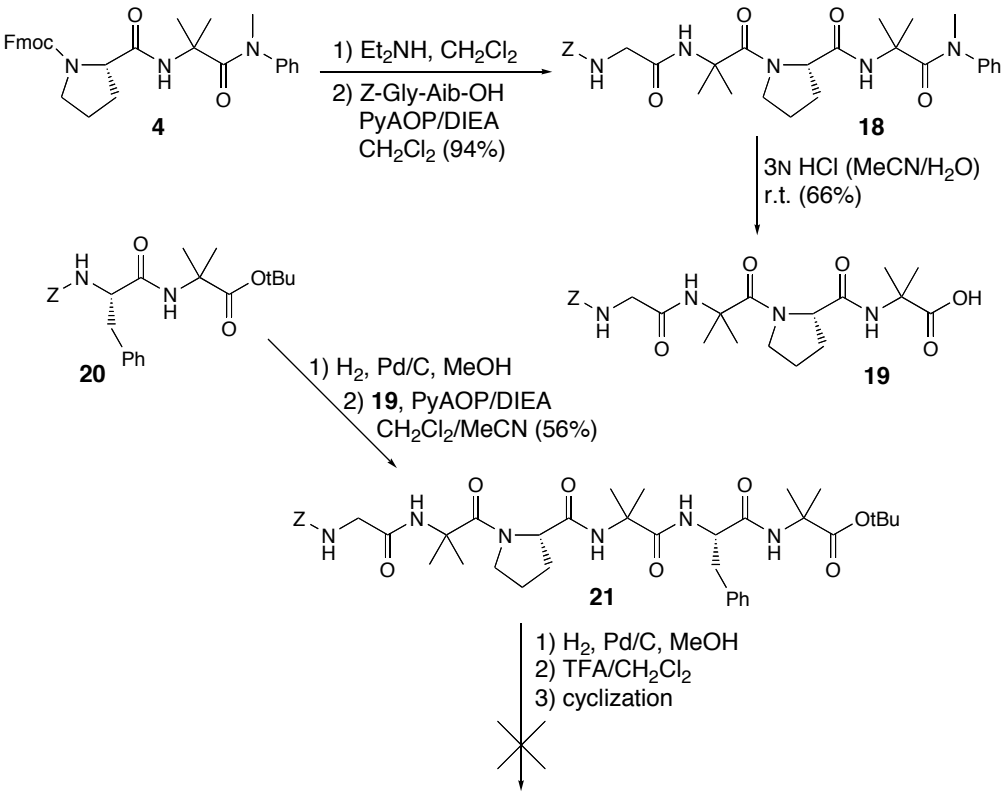
cyclo(Pro-Aib-Gly-Aib-Aib-Phe)

1

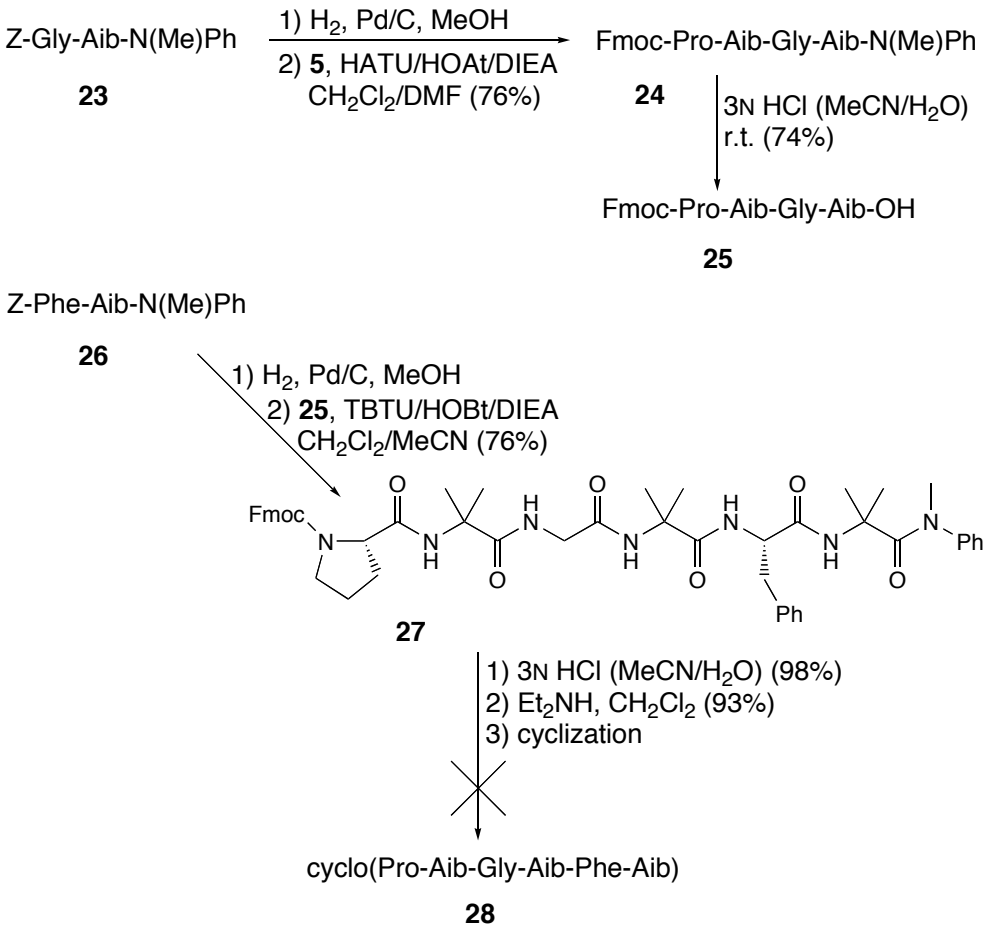
Scheme 4


Z
Ph
6
1) H2, Pd/C
2) Fmoc-Pro-OH
PyAOP/DIEA
CH2Cl2 (93%)
Fmoc
15
1) Et2NH, CH2Cl2 (82%)
2) Z-Gly-Aib-OH, PyAOP/DIEA
CH2Cl2 (87%)
16 X = N(Me)Ph
17 X = OH
3N HCl
(MeCN/H2O)
r.t. (82%)
1) H2, Pd/C, MeOH
2) DEPC, DIEA, DMF
(36%)
cyclo(Gly-Aib-Pro-Gly-Aib-Aib)
2

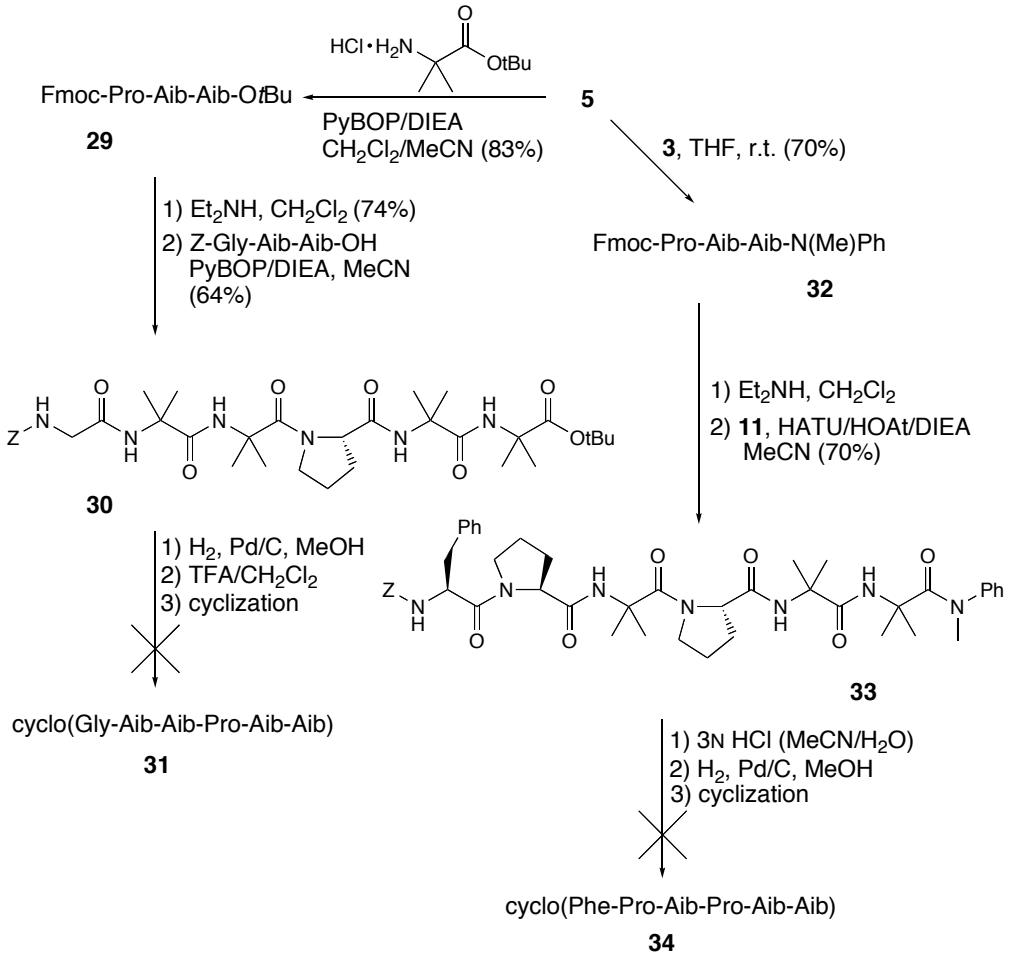
Scheme 5


Fmoc
Ph
4
1) Et2NH, CH2Cl2
2) Z-Gly-Aib-OH
PyAOP/DIEA
CH2Cl2 (94%)
Z
18
3N HCl (MeCN/H2O)
r.t. (66%)
OtBu
20
Ph
19
OH
1) H2, Pd/C, MeOH
2) 19, PyAOP/DIEA
CH2Cl2/MeCN (56%)
21
1) H2, Pd/C, MeOH
2) TFA/CH2Cl2
3) cyclization
cyclo(Gly-Aib-Pro-Aib-Phe-Aib)
22

Scheme 6


Z-Gly-Aib-N(Me)Ph
23
1) H2, Pd/C, MeOH
2) 5, HATU/HOAt/DIEA
CH2Cl2/DMF (76%)
Fmoc-Pro-Aib-Gly-Aib-N(Me)Ph
24
3N HCl (MeCN/H2O)
r.t. (74%)
Fmoc-Pro-Aib-Gly-Aib-OH
25
Z-Phe-Aib-N(Me)Ph
26
1) H2, Pd/C, MeOH
2) 25, TBTU/HOBt/DIEA
CH2Cl2/MeCN (76%)
Fmoc
Ph
27
1) 3N HCl (MeCN/H2O) (98%)
2) Et2NH, CH2Cl2 (93%)
3) cyclization
cyclo(Pro-Aib-Gly-Aib-Phe-Aib)
28

Scheme 7


HCl • H2N
OtBu
Fmoc-Pro-Aib-Aib-OtBu
29
PyBOP/DIEA
CH2Cl2/MeCN (83%)
5
3, THF, r.t. (70%)
1) Et2NH, CH2Cl2 (74%)
2) Z-Gly-Aib-Aib-OH
PyBOP/DIEA, MeCN
(64%)
Fmoc-Pro-Aib-Aib-N(Me)Ph
32
1) Et2NH, CH2Cl2
2) 11, HATU/HOAt/DIEA
MeCN (70%)
30
1) H2, Pd/C, MeOH
2) TFA/CH2Cl2
3) cyclization
33
1) 3N HCl (MeCN/H2O)
2) H2, Pd/C, MeOH
3) cyclization
cyclo(Gly-Aib-Aib-Pro-Aib-Aib)
31
cyclo(Phe-Pro-Aib-Pro-Aib-Aib)
34

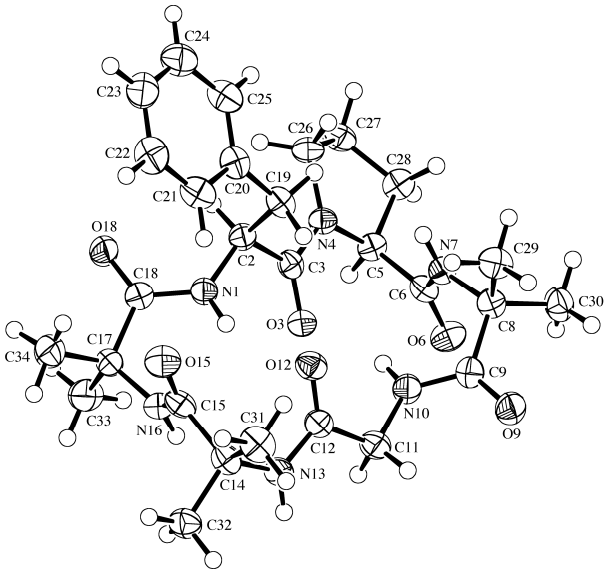


Fig. 1. *ORTEP Plot* [21] *of the molecular structure of* **1**. *The co-crystallized* Et(*i*-Pr)$_2$NH$^+$PF$_6^-$ *and* H_2O *are not shown* (50% probability ellipsoids, arbitrary numbering of the atoms)